ISSN 1821-1046
UDK 630

INSTITUTE OF FORESTRY
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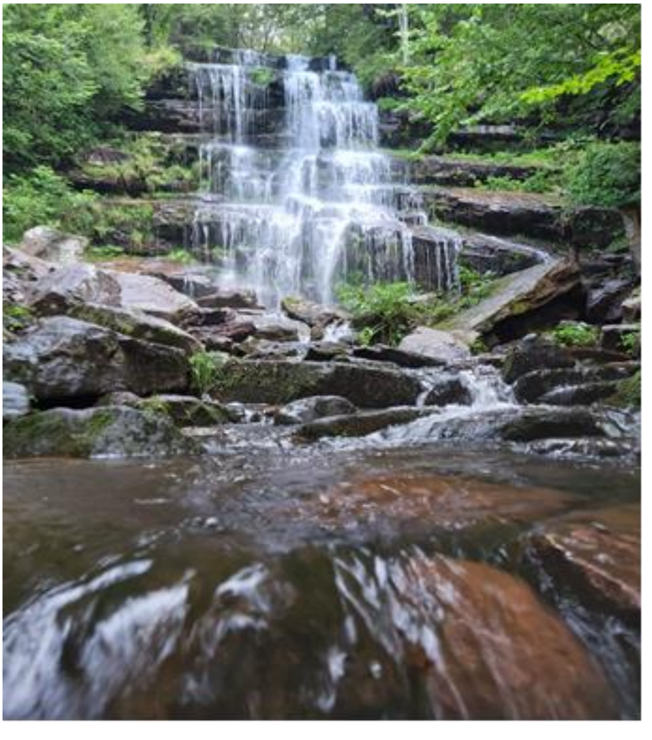
SUSTAINABLE FORESTRY ODRŽIVO ŠUMARSTVO

COLLECTION
Vol. 85-86

ZBORNIK RADOVA
Vol. 85-86



BELGRADE BEOGRAD
2022.

ISSN 1821-1046
9 771821 104000

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INSTITUTE OF FORESTRY BELGRADE	**INSTITUT ZA ŠUMARSTVO BEOGRAD**
COLLECTION OF PAPERS	**ZBORNIK RADOVA**
Publisher	**Izdavač**
Institute of Forestry Belgrade, Serbia	Institut za šumarstvo Beograd, Srbija
For Publisher	**Za izdavača**
Ljubinko Rakonjac, Ph.D.	Dr Ljubinko Rakonjac
Editor-in-Chief	**Glavni i odgovorni urednik**
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Printed in	**Tiraž**
100 copies	100 primeraka

Printed by	**Štampa**
Black and White Belgrade	Black and White Beograd

Cover Page: Author of the Photos, B.Sc. Nenad Šurjanac
Locality: Stara Planina

Naslovna strana: Autor fotografije Nenad Šurjanac, dipl. inž.
Lokalitet: Stara planina

CONTENT SADRŽAJ

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INSTITUTE OF FORESTRY • BELGRADE
INSTITUT ZA ŠUMARSTVO • BEOGRAD

SUSTAINABLE FORESTRY COLLECTION 85-86, 2022
ODRŽIVO ŠUMARSTVO ZBORNIK RADOVA 85-86, 2022

DOI: 10.5937/SustFor2285035M
Original scientific paper

MORPHOMETRIC CHARACTERISTICS OF *PAULOWNIA ELONGATA* S. Y. HU. AND *PAULOWNIA FORTUNEI* SEEM. HEMSL. LEAVES AND FERTILISATION IN DIFFERENT SITES

Suzana MITROVIĆ[1], Milorad VESELINOVIĆ[1], Nevena ČULE[1], Goran ČEŠLJAR[1], Saša EREMIJA[1], Renata GAGIĆ-SERDAR[1], Snežana STAJIĆ[1]

Abstract: *The paper presents the results of the analysis dealing with the impact of plant fertilisation in the first year after planting on the leaf morphological characteristics. The analysis was conducted within the research into the potential introduction and adaptation of paulownia to different sites in Serbia. The results related to the effects of fertilisation on the quality of plant leaves are useful for the cultivation of certain types of soil where the morphometric analysis of leaves reveals structure-function relationships, i.e., more detailed indicators of the species' adaptability. The research was conducted in two localities. Sample plots with Paulownia elongata S. Y. Hu. and Paulownia fortunei Seem. Hemsl. were established in Obrenovac and Pambukovica, where leaf material was collected for laboratory analysis. The following morphometric leaf characteristics were measured: leaf area, leaf perimeter, leaf lamina length, central nerve length, maximum leaf width, leaf width at 1 cm from the base of the leaf, petiole length, distance between the 3rd and 4th nerve, the number of nerves to the left side of the midrib, and the number of veins to the right side of the midrib. The obtained results of leaf morphometric measurements were statistically processed in the Statgraphics software. Based on the results of measuring the leaf morphometric characteristics, fertilisation has a positive effect on the size of the leaves of the studied paulownia species.*

[1]Ph.D. Suzana Mitrović, Senior Research Associate, Ph.D. Milorad Veselinović, Principal Research Fellow, Ph.D. Nevena Čule, Senior Research Associate, Ph.D. Goran Češljar, Research Associate, Ph.D. Saša Eremija, Senior Research Associate, Ph.D. Renata Gagić, Research Associate, Ph.D. Snežana Stajić, Senior Research Associate, Institute of Forestry, Belgrade, Serbia

Corresponding author: Suzana Mitrović, e-mail*: suzana.mitrovic@forest.org.rs*

Key words: fast-growing species, leaf morphology, phenotypic stability, introduction, adaptation.

MORFOMETRIJSKE KARAKTERISTIKI LISTOVA VRSTA *PAULOWNIA ELONGATA* S. Y. HU. I *PAULOWNIA FORTUNEI* SEEM. HEMSL. U ODNOSU NA PRIHRANJIVANJE NA RAZLIČITIM STANIŠTIMA

Izvod: *U radu su prikazani rezultati analize uticaja prihranjivanja biljaka u prvoj godini nakon sadnje, na morfološke karakteristike listova u okviru istraživanje mogućnosti introdukcije i adaptacije paulovnija na različita staništa u Srbiji. Dobijanje rezultata o uticaju prihranjivanja na kvalitet listova biljaka je značajno za gajenje na određenim tipovima zemljišta gde morfometrijska analiza listova pokazuje strukturno-funkcionalne veze, odnosno detaljnije pokazatelje adaptibilnost vrste. Istraživanja su sprovedena na dva lokaliteta. Ogledna polja na lokalitetima u Obrenovcu i Pambukovici su osnovana sa vrstama Paulownia elongata S. Y. Hu. i Paulownia fortunei Seem. Hemsl.. U okviru oglednih polja vršeno je prikupljanje lisnog materijala za analizu u laboratorijskim uslovima. Izvršena su merenja morfometrijskih karakteristika listova: površina lista, obim lista, dužina lisne ploče, dužina centralnog nerva, širina lista na najširem delu lisne ploče, širina lista na 1 cm od osnove lista, dužina peteljke, razmak između 3. i 4. nerva, broj nerava na levoj strani od centralnog nerva, i broj nerava na desnoj strani od centralnog nerva. Dobijeni rezultati morfometrijskih merenja listova statistički su obrađeni u programu Statgraphics. Na osnovu rezultata merenja morfometrijskih karakteristika listova utvrđeno je da prihranjivanja ima pozitivan uticaj na veličinu listova analiziranih vrsta paulovnije.*

Ključne reči: brzorastuće vrste, morfologija lista, fenotipska stabilnost, introdukcija, adaptacija.

1. INTRODUCTION

Besides efforts to preserve and protect native species, modern society is faced with the need to introduce new species that are resilient to changing environmental conditions and human impact. Preserving and increasing areas under forests is a high priority while selecting adequate species poses serious challenges (Ivetić and Vilotić, 2014). Numerous research studies deal with the introduction of new species and their adaptation to ongoing climatic and environmental changes. These studies are of great importance in finding the most effective solutions to reduce the adverse effects of climate change (Betts et al., 2008; Innes et al., 2009). The limited supply and increasing demand for wood have made the use of fast-growing species for wood and biomass production a pressing need (Swamy et al., 2006; Mishra et al., 2010).

In the struggle to enhance plantation forestry in Serbia, the potential use of new tree species has to be given greater focus (Ivetić and Vilotić, 2014). Due to its rapid growth and coppice vigour, Paulownia Sieb. & Zucc. species have high biomass and biofuel production potential (Lucas-Borja et al., 2011; Yadav et al., 2013).

Thanks to the large leaf mass rich in nitrites, Paulownia litterfall has a crucial ameliorative role in improving the soil quality around trees. Because of this property, in some areas in China, green manure is made with Paulownia leaves and used to improve the ecological properties of the soil (Zhu et al., 1986). When mixing it with other tree species, it is necessary to follow the growth trends because, as a light-loving species, Paulownia is unsuitable for mixing with other similar species (Zhu et al., 1986; Williams, 1993).

Afforestation is usually carried out in soils poor in nutrients that have to be improved by adding different types of fertilisers (Stilinović, 1991). The decision on when what and how much fertiliser to use depends primarily on the soil conditions. It is also affected by biological characteristics and the stage in the development of plants (Tucović and Simić, 2002; Hawkins et al., 2005). Providing plants with a proper nutrition system or necessary plant assimilates should ensure the formation of biologically healthy material resistant to adverse environmental conditions. This way, plants can develop and grow well in plantations and cultures (Jacobs et al., 2005, Mitrović et al., 2012). Research by García-Morote et al. (2014) show that fast-growing species can be grown even in semi-arid regions, provided they obtain minimum irrigation.

In young Paulownia trees (in the juvenile development phase), the leaves have a wavy rim with pronounced lobes. They are extremely large and can be as long as 90 cm (Graves, 1989). On the other hand, the leaves of mature trees are smaller, 15-30 cm in length and 10-20 cm in width, full perimeter (Innes, 2009). The leaves are covered with thick hairs on both sides (Kays et al., 1998).

The large and hairy Paulownia leaves have a very useful role in removing sulfur and carbon dioxide from the air and retaining solid pollutants, smoke and dust in the canopy (Zhu et al., 1986; Šijačić-Nikolić et al., 2008). Zhang et al. (2007) investigated contaminated soil near a lead and zinc smelter. The obtained results suggest that seedlings of Paulownia fortunei Seem. Hemsl. growing on soil highly contaminated with heavy metals can significantly improve the structure and function of the communities of soil microorganisms. Paulownia is considered a pioneer species (Zhu et al., 1986). It can accumulate large amounts of heavy metals and thus perform soil phytoremediation (Stanković et al., 2009; Bahri et al., 2015). The content of heavy metals in the leaves of Paulownia growing in urban conditions shows that this genus has a high urban pollution tolerance.

All these facts indicate that Paulownia trees can be used in the restoration of degraded land and wastewater, and grown in tree rows, windbreaks, urban environments and roadsides (Doumett et al., 2011, Mitrović et al., 2011).

The size of the leaf is vital for the process of photosynthesis and the production of nutrients that are directly correlated with the production of the entire plant biomass and the role of leaves in air purification. Therefore we have conducted a comparative analysis of ten main leaf properties (Morphological leaf analysis was done according to the modified protocol “Assessments of oak leaf morphology“) and plant fertilisation in the first year after planting.

2. MATERIAL AND METHODS

Sample plots were established in localities with different orographic features, climatic conditions and physical and chemical properties of the soil:

1. The sample plot in Veliko Polje near Obrenovac – Site 1;
2. The sample plot in the village of Pambukovica near Ub – Site 2;

The sample plots at the sites in Obrenovac (Site 1) and Pambukovica (Site 2) were established by planting Paulownia trees of generative origin. The seeds were collected from well-adapted genotypes of two Paulownia species from a sample area in Bela Crkva. Container seedlings produced by generative reproduction were used as starting material to establish experimental plantations.

Seedlings were planted in rows at a 4x4 m spacing. Each row had 25 plants. There were 12 rows of *Paulownia elongata* seedlings and 12 seedlings of *Paulownia fortunei* species. The seedlings were planted by hand in holes with a diameter of 30x30 cm.

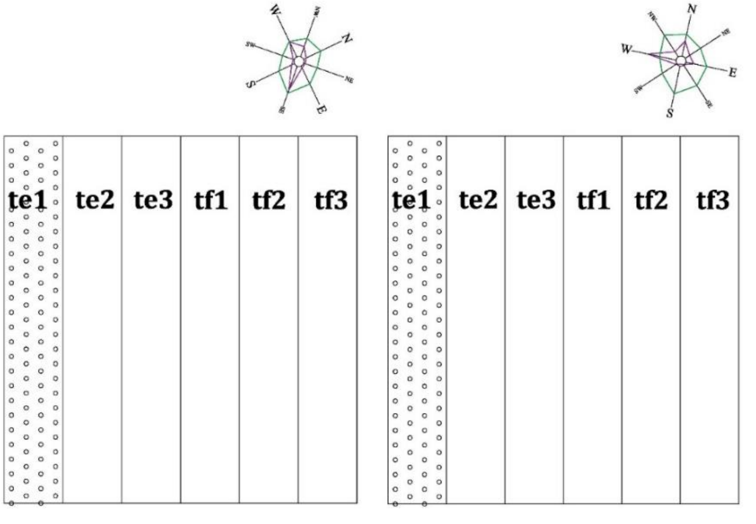

Figure 1. *Schema of sample plots with a wind rose (velocity and frequency) in Obrenovac (left) and Pambukovica (right)*

(te1 – seedlings of P. elongata from treatment group 1 (with a large amount of fertiliser added); te2 – seedlings of P. elongata from treatment group 2 (with a small amount of fertiliser added); te3 – seedlings of P. elongata not fertilised (control group); tf1 – seedlings of P fortunei from treatment group 1 (with a large amount of fertiliser added); tf2 – P. fortunei seedlings from treatment group 2 (with a small amount of fertiliser added); tf3 - P. fortunei seedlings not fertilised (control group)

Each sample plot was divided into six treatment groups (each treatment group had four rows of 25 plants), which differed in the amount of fertiliser (fertor) added and the control group of plants that were not fertilised.

The type of fertiliser to be used to supply plants with nutrients was selected based on soil analysis. Due to the heavy mechanical composition and the acidic pH, chicken manure – fertor (http://www.mrf-garden.com) was used in both localities. Fertor is an organic fertiliser (in the form of pellets) produced from 100% chicken manure, with other organic substances of plant origin added to increase its nutritional value. A high share of total organic matter maintains and improves the physical properties of the soil. Some macro- and micro-elements are easily accessible and available to plants, while some are released gradually. The fertiliser was added to the plants in the amount of 240 g per plant (t1) and 120 g per plant (t2). Control areas (t3) did not receive any fertiliser.

Leaves were sampled in the field at the end of the first and second growing seasons. They were collected from the same portion of the canopy, i.e., the same nodes.

The collected leaves were herbilised and scanned. Using the AutoCAD software, the scanned leaves were measured with a precision of 1 mm. The sample included 150 plants per site. Five leaves were taken from each plant whose morphometric characteristics were to be measured. It added up to 750 leaves per site or 1500 for both.

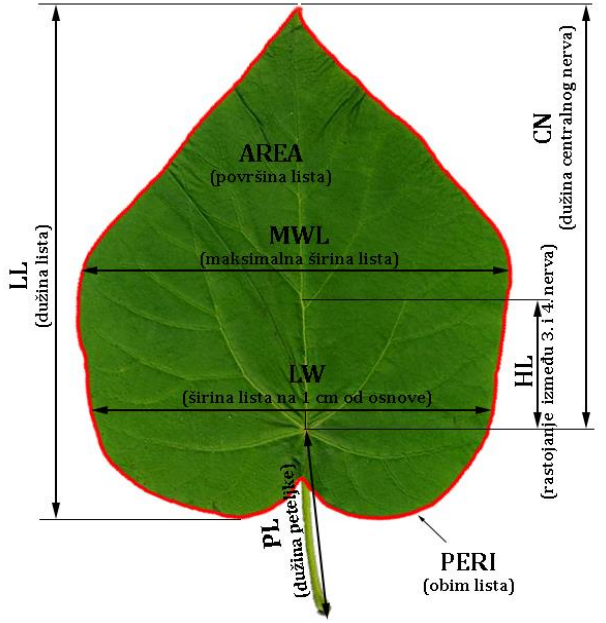

Figure 2. *Schematic representation of analysed measurement parameters of Paulownia leaves*

Source:http://www.european-trees.com/requete-leaf-simple-lisse--1a4.html (modified)

Ten following ten leaf properties were measured (the morphological leaf assessment was done according to the modified protocol of "Assessments of oak leaf morphology": http:/www.pierroton.inra.fr/Fairoak/Protocoles/OAKMORPH.html):

1. total leaf area without petiole, in cm^2 (AREA);
2. total leaf perimeter, without petiole, in cm (PERI);

3. length of the leaf lamina, from the leaf base to the top of the leaf, in cm (LL);
4. length of the central nerve (CN);
5. maximum leaf width, in cm (MWL);
6. leaf width at 1 cm from the leaf base, in cm (LW);
7. leaf petiole length, from the base of the leaf to the top of the petiole, in cm (PL);
8. distance between the 3rd and 4th nerve, in cm (HL);
9. number of nerves on the left side of the leaf (NLL); and
10. number of nerves on the right side of the leaf (NLR).

Special data entry forms were created for leaf morphometric measurements. They were statistically processed in the STATGRAPHICS software (Statistical Graphics Corporation, USA). The experimental design corresponds to a three-way and two-way analysis of variance: ANOVA III.

- factor A (site) with 2 levels: Site 1 (Obrenovac) and Site 2 (Pambukovica);
- factor B (species) with 2 levels: species 1 (*P. elongata*) and species 2 (*P. fortunei*);
- factor C (treatment groups) with 3 levels: treatment 1, treatment 2 and treatment 3.

ANOVA III assesses the effect of each factor separately and their interactions. The size of the aggregate sample (number of aggregate sample elements) for ANOVA III is n=1500 (2x2x3x125=1500) within which the following ten properties were measured: leaf area, leaf perimeter, leaf lamina length, central nerve length, maximum leaf width, leaf width at 1 cm from the leaf base, leaf petiole length, distance between the 3rd and 4th nerve, the number of nerves to the left of the leaf nerve, and the number of nerves to the right of the central leaf nerve.

4. RESULTS AND DISCUSSION

Site 1 is located in Veliko Polje, Obrenovac municipality. The sample plot is located on the left bank of the Kolubara River at 74 m a.s.l. The terrain of the plot is flat without a slope, and the plant rows are northwest-southeast oriented.

Table 1. *Physical properties of the soil at the site in Obrenovac (Site 1)*

Profile depth (cm)	Coarse sand	Fine sand	Silt	Clay	Total sand	Total clay	Texture class
	%						
0-20	4.50	48.62	20.58	26.30	53.12	46.88	Sandy loam
20-40	4.20	46.60	22.30	26.90	50.80	49.20	Loam

The soil at Site 1 has good water and air permeability and a sufficiently high retention capacity of available water. Regarding its texture, it belongs to the class of sandy loam. The surface soil layer belongs to the class of sandy loam, and the deeper layer to loam. Although the analysed layers belong to different textural classes, they are similar, i.e., there is no strong differentiation of profiles by textural composition. The content of individual textural fractions in both layers is close to the limit values between sandy loam and loam. This soil is well-permeable to water and well-aerated throughout the depth of the solum (Table 1).

Table 2. *Chemical properties of the soil at the site in Obrenovac (Site 1)*

Profile depth (cm)	pH		Adsorptive complex					Total		C/N	Available	
	H2O	KCl	T	S	T-S	V	Y1	humus	N		P2O5	K2O
			equ.m.mol/100g			%	cm^3	%	%		mg/100g	
0-20	5.91	4.74	32.50	24.94	7.56	76.74	11.63	2.55	0.19	7.59	23.55	19.39
20-40	5.87	4.77	32.90	25.27	7.63	76.80	11.74	2.03	0.12	10.13	21.63	17.45

Site 1 has medium soil pH. The total adsorption capacity is high due to the high proportion of clay in the texture. According to the content of total humus, both analysed layers are low in humus. A narrow C/N ratio is favourable for the mineralisation of organic matter. The availability of easily accessible forms of phosphorus is good throughout the solum depth, and the availability of accessible potassium is medium (Table 2).

Site 2 is located in Pambukovica near Ub. It belongs to the Kolubara District in the Tamnava microregion. The sample plot is located on Jastrebovac Hill, municipality of Ub. The plot has undulating terrain with the lowest point at 162.60 m and the highest at 176.11 m above sea level. The relief of this area is flat to hilly, with slight height differences. The plot mostly has a southeastern aspect. It is northwestern in a small part. The plot is surrounded by beech-oak forests. The average altitude is 174.69 m. The planted rows run north to south.

Table 3. *Physical properties of the soil at the site in Pambukovica (Site 2)*

Profile depth (cm)	Coarse sand	Fine sand	Silt	Clay	Total		Texture class
					sand	clay	
	%						
0-20	0.60	44.50	29.00	25.90	45.10	54.90	Loam
20-40	0.40	42.20	25.70	31.70	42.60	57.40	Sandy clay loam

The surface layer of the soil at Site 2 belongs to the loam class and has good water and air permeability. With the depth of the soil solum, the share of clay and fine sand increases, and the textural class changes to sandy clay loam, which is slightly less permeable to water and air (Table 3).

Table 4. *Chemical properties of the soil at the site in Pambukovica (Site 2)*

Profile depth (cm)	pH		Adsorptive complex					Total		C/N	Available	
	H2O	KCl	T	S	T-S	V	Y1	humus	N		P2O5	K2O
			equ.m.mol/100g			%	cm^3	%	%		mg/100g	
0-20	5.35	3.84	32.13	19.56	12.57	60.87	19.34	1.47	0.15	5.87	<LD	8.76
20-40	5.41	3.94	32.66	21.51	11.15	65.86	17.15	0.98	0.13	4.41	<LD	7.94

The soil pH range is very acidic throughout the whole solum. The total adsorption capacity is high. The surface layer has a low level of total humus content, while the deeper layer has a low to very low level. The content of total nitrogen is low, and the ratio of carbon to nitrogen is narrow. In both layers we studied, the amount of plant-available phosphorus is below the detection limit of the Al-method, which means that this soil is very poor in plant-available forms of phosphorus. The amounts of plant-available forms of potassium are within the limits of poor availability throughout the entire depth of the profile (Table 4).

Table 5 and Table 6 present the mean values of the analysed leaf parameters of seedlings at both sites, obtained by descriptive statistics.

Regarding the property **leaf area** (Table 5), there are differences in the mean values of the groups (measurements) depending on site, species and treatment factors. Seedlings at Site 1 have significantly higher mean values of leaf area than seedlings at Site 2. The difference between species is statistically significant, i.e., *Paulownia elongata* seedlings have higher mean values of leaf area compared to *Paulownia fortunei* seedlings. The difference between all three treatments is statistically significant. The seedlings from the group fed with a large amount of fertiliser have higher mean values of the leaf area, and the seedlings from the control group have the lowest mean values.

Regarding the property **leaf perimeter** (Table 5), there are differences in the mean values of the groups (measurements) for site, species and treatment factors. Seedlings at Site 1 show significantly higher values of leaf perimeter than seedlings at Site 2. The difference in the mean leaf perimeter values between species is statistically significant. Seedlings of *Paulownia elongata* show significantly higher means. The difference in the mean values of leaf perimeter between seedlings of different treatments is also statistically significant. Seedlings of the group fed with a large amount of fertiliser have the highest mean values and the lowest in the control group. Interactions between all three examined factors are not statistically significant.

For the property **leaf lamina length** (Table 5), the differences in the mean values of the groups (measurements) depend on the site and treatment, but not the species. Mean values of leaf lamina length differ significantly between these two sites, with the seedlings at Site 1 showing higher mean values. There is no statistically significant difference between the species in the mean value of leaf lamina length, although seedlings of *Paulownia elongata* have higher mean values. The differences in mean values between the seedlings of different treatment groups are also statistically significant, with the highest mean value of the leaf lamina length achieved by the seedlings from the group that was treated with a large amount of fertiliser, followed by the group that was treated with a small amount of fertiliser. The lowest mean value was attained by the seedlings from the control group (Table 5).

The property **central nerve length** (Table 5) shows differences in the mean values of the groups (measurements) for site and treatment factors, but not species. Seedlings of Site 1 have significantly higher mean values of the central nerve length. The difference in the mean value of the central nerve length between species is not statistically significant, although *Paulownia elongata* seedlings have higher mean values. Seedlings in the group that was treated with a large amount of fertiliser have significantly higher mean values than seedlings from the group treated with a small amount of fertiliser and the control group. Seedlings within the latter two groups do not differ significantly, although seedlings in treatment group 2 have higher mean values of the central nerve length than control seedlings. (Table 5).

Regarding the **leaf width** (Table 5), there are differences in the mean values of the groups (measurements) for site and treatment factors. As with previous properties, the species factor does not lead to any differences in the mean values of the groups (measurements). At both sites, there are significant differences between the seedlings in the mean value of leaf width which is higher at Site 1. The difference in the mean values of the property between species is not statistically significant, although seedlings of *Paulownia elongata* have higher mean values. Mean values of leaf width are significantly different between seedlings of different treatment groups. The seedlings treated with a large amount of fertiliser have the highest mean value of the leaf width property, while the control group has the lowest. (Table 5).

Regarding the **leaf width at 1 cm from the base** (Table 6), there are differences in the mean values of the groups (measurements) for site and treatment factors, but not species. There is a significant difference in the mean value of leaf width at 1 cm from the base between the seedlings at different sites. It is significantly higher in the seedlings at Site 1. *Paulownia elongata* seedlings have larger (7.24 cm) mean leaf width values than *Paulownia fortunei* seedlings, but the difference is not statistically significant. Seedlings of different treatment groups have significantly different mean values of leaf width at 1 cm from the base, whereby seedlings treated with a large amount of fertiliser have significantly higher mean values of the property compared to seedlings treated with less fertiliser and control treatment (Table 6).

Table 5. *Basic descriptive statistics parameters and three-way ANOVA for leaf properties: leaf area (cm^2), leaf perimeter (cm), leaf lamina length (cm), central nerve length (cm), leaf width (cm); for seedlings at the site in Obrenovac (Site 1) and Pambukovica (Site 2) at the end of the first growing season*

factor	level	leaf area	leaf perimeter	leaf lamina length	central nerve length	leaf width
Site (A)	Site 1	[A] 57,25(45,14)^{b}	31,26(20,01)^{b}	9,30(4,50)^{b}	7,81(3,56)^{b}	8,51(3,97)^{b}
	Site 2	38,02(19,17)^{a}	24,84(13,70)^{a}	7,48(2,33)^{a}	6,12(1,90)^{a}	6,82(2,28)^{a}
		[B] $F_{1,1490}$=124,71*	$F_{1,1490}$=54,53*	$F_{1,1490}$=105,47*	$F_{1,1490}$=146,68*	$F_{1,1490}$=109,94*
Species (B)	*P. elongata*	50,06(41,50)^{b}	29,19(16,64)^{b}	8,53(4,18)	7,00(3,35)	7,75(3,06)
	P. fortunei	45,22(29,28)^{a}	26,91(18,15)^{a}	8,25(3,13)	6,93(2,54)	7,58(3,60)
		$F_{1,1490}$=8,04*	$F_{1,1490}$=7,00*	ns	ns	ns
Treatment (C)	treatment 1	57,57(35,53)^{c}	31,81(16,18)^{c}	9,28(3,01)^{c}	7,64(2,53)^{b}	8,44(3,81)^{c}
	treatment 2	49,30(45,38)^{b}	28,25(15,64)^{b}	8,29(3,11)^{b}	6,74(2,58)^{a}	7,74(3,46)^{b}
	treatment 3	36,02(18,22)^{a}	24,08(19,42)^{a}	7,60(4,56)^{a}	6,47(3,56)^{a}	6,82(2,41)^{a}
		$F_{2,1490}$=53,13*	$F_{2,1490}$=26,38*	$F_{2,1490}$=30,15*	$F_{2,1490}$=26,99*	$F_{2,1490}$=33,62*
interactions **(AXB)**		ns	ns	ns	ns	ns
interactions **(AXC)**		$F_{2,1490}$=7,69*	ns	ns	ns	$F_{2,1490}$=4,89*
interactions **(BXC)**		ns	ns	$F_{2,1490}$=32,13*	$F_{2,1490}$=57,34*	$F_{2,1490}$=18,87*

Table 6. *Basic descriptive statistics parameters and three-way ANOVA for leaf properties: leaf width at 1 cm from the base (cm), petiole length (cm), distance between the third and fourth nerve, number of nerves - left and number of nerves - right for seedlings at the site in Obrenovac (Site 1) and Pambukovica (Site 2) at the end of the first growing season*

factor	level	leaf width at 1 cm from the base	petiole length	distance between the third and fourth nerve	number of nerves - left	number of nerves - right
Site (A)	Site I	7.74(3.06)^{b}	6.38(3.22)^{b}	2.11(0.98)^{b}	8.49(1.04)^{b}	8.54(0.98)^{b}
	Site II	6.53(4.16)^{a}	4.57(1.73)^{a}	1.41(0.58)^{a}	8.27(0.91)^{a}	8.19(0.78)^{a}
		$F_{1.1490}$=42.48*	$F_{1.1490}$=223.56*	$F_{1.1490}$=319.79*	$F_{1.1490}$=24.23*	$F_{1.1490}$=74.99*
Species (B)	*P. elongata*	7.24(4.01)	5.88(2.79)^{b}	1.74(0.92)	8.51(1.02)^{b}	8.44(0.97)^{b}
	P. fortunei	7.03(3.36)	5.07(2.62)^{a}	1.78(0.84)	8.24(0.93)^{a}	8.30(0.84)^{a}

factor	level	leaf width at 1 cm from the base	petiole length	distance between the third and fourth nerve	number of nerves - left	number of nerves - right
		ns	$F_{1.1490}$=46.68*	ns	$F_{1.1490}$=36.60*	$F_{1.1490}$=11.88*
Treatment (C)	treatment 1	7.88(4.58)[c]	6.37(2.90)[c]	2.00(0.87)[c]	8.45(1.15)	8.46(1.08)[b]
	treatment 2	7.19(2.94)[b]	5.29(2.88)[b]	1.75(0.81)[b]	8.35(0.79)	8.34(0.81)[a]
	treatment	6.32(3.21)[a]	4.76(2.10)[a]	1.53(0.89)[a]	8.34(0.98)	8.31(0.80)[a]
		$F_{2.1490}$=23.71*	$F_{2.1490}$=61.76*	$F_{2.1490}$=46.27*	ns	$F_{2.1490}$=5.08*
***interactions* (AXB)**		ns	$F_{1.1490}$=28.58*	$F_{1.1490}$=8.01*	$F_{1.1490}$=132.14*	$F_{1.1490}$=103.36*
***interactions* (AXC)**		ns	$F_{2.1490}$=20.26*	$F_{2.1490}$=7.65*	$F_{2.1490}$=22.61*	$F_{2.1490}$=25.10*
***interactions* (BXC)**		$F_{2.1490}$=8.21*	$F_{2.1490}$=57.52*	$F_{2.1490}$=32.08*	$F_{2.1490}$=96.22*	$F_{2.1490}$=124.15*

* Three-way analysis of variance (ANOVA III). Factor A (site) with 2 levels: Site 1 (Obrenovac) and Site 2 (Pambukovica); factor B (species) with 2 levels: species 1 (*P. elongata*) and species 2 (*P. fortunei*); factor C (treatment) with 3 levels: treatment 1 (high amount of fertiliser), treatment 2 (low amount of fertiliser), and treatment 3 (control), and their interactions. Aggregate sample size (number of aggregate sample elements), n=1500 (2 sites x 2 species x 3 treatments x 125 = 1500). [A] =mean value (standard deviation); [B]= F-test indicator with numbers of degrees of freedom; ns = non-significant difference between the mean values of the populations (P> 0.05); * = statistically significant difference ($P\leq$ 0.05).

As for the **petiole length** (Table 6), there are differences in the mean values of the groups (measurements) for the location, species and treatment factors. Seedlings at Site 1 have significantly higher mean values of petiole length. Seedlings of *Paulownia elongata* have significantly higher mean values of petiole length. The difference in the mean value of the petiole length between the seedlings with different treatments is significant. The mean value of the petiole length is the highest in the seedlings treated with a large amount of fertiliser and the smallest in the seedlings from the control group.

For the **distance between the third and fourth nerve of the leaf** (Table 6), there are differences in the mean values of the groups (measurements) for the site and treatment factors, but not species. Seedlings at Site 1 show significantly higher values of the distance between the third and fourth nerve of the leaf. There is no statistically significant difference between the species in the mean value of this property, although seedlings of the *Paulownia fortunei* have higher mean values. There is a statistically significant difference in the mean values of the distance between the third and fourth nerves between the seedlings of different treatment groups. Seedlings from the group treated with a large amount of fertiliser show the highest mean values and the control treatment group the lowest.

Regarding the **number of nerves on the left side of the leaf** (Table 6), the mean values of the groups (measurements) differ depending on the site and species factors, but not treatment. Seedlings at Site 1 have significantly higher mean values of the number of nerves on the left side of the leaf. *Paulownia elongata* seedlings have significantly higher mean values of the number of nerves on the left side of the leaf. There is no statistically significant difference in the number of nerves between seedlings of different treatments. The highest mean values of the number of nerves on the left side of the leaf are shown by seedlings from the group treated with a large amount of fertiliser and the least by the control group.

As for the **number of nerves on the right side of the leaf** (Table 6), there are differences in the mean values of the groups (measurements) depending on the site, species and treatment factors. Seedlings at Site 1 have a significantly higher mean number of nerves on the right side of the leaf. *Paulownia elongata* seedlings have significantly higher mean values of the number of nerves on the right side. Between seedlings in different treatment groups, the difference in the mean value of the number of nerves on the right side of the leaf is significant. Seedlings from the group treated with a large amount of fertiliser have significantly higher mean values of the number of nerves than seedlings from the group treated with a small amount of fertiliser and from the control group (8.31) which are not significantly different from each other.

The results of the analysis of variance in the first growing season reveal significant differences in the morphometric characteristics of the leaves between the seedlings at both sites (Table 5 and Table 6). The most significant difference is between the parameters that determine the leaf size. The mean value of the leaf lamina length of the seedlings at Site 1 is 9.30 cm and 7.48 cm at Site 2. The mean values of petiole length are 6.38 cm and 4.57 cm, respectively. The leaf size achieved by plants at both sites is far below the values that Paulownia can reach (Graves, 1989; Šijačić-Nikolić, et al., 2009). The shape and structure of leaves depend significantly on a range of factors (Fender et al., 2011; Stojnić, 2013). The morphometric characteristics of the seedling leaves at both sites are a result of their adaptation to the unfavourable environmental conditions in which the seedlings have grown. At Site 2, the content of nitrogen, phosphorus and potassium in the soil (also reflected on the leaves) is lower than at Site I, which makes the conditions for plant growth and leaf development less favourable. Although the average values of precipitation in the growing season were higher at Site 2, the seedlings did not have enough moisture in the soil, because atmospheric precipitation quickly ran off from the sloping plot, and the leaves were consequently smaller. In their research, Ozturk et al. (2014) state that the size of leaves depends on climatic factors and are smaller in drier environmental conditions (Pedrol et al., 2000; Otieno et al., 2005).

There are differences in the leaf morphometric characteristics between the seedlings of the analysed species, as indicated in literature (Šilić, 1990; Cvjetićanin and Perović, 2009), whereby the *Paulownia elongata* seedlings have higher mean values of all parameters compared to the *Paulownia fortunei* seedlings at both sites.

The effects of fertilisation can be seen in differences between leaf morphometric parameters. Seedlings that are fed with a large amount of fertiliser have the highest mean values. These findings are in agreement with the results obtained by Boughalleb et al. (2011) and Adejobi et al. (2014) that feeding had a positive effect on leaf size.

5. CONCLUSIONS

The analysis of leaf morphological parameters of *Paulownia elongata* S. Y. Hu. and *Paulownia fortunei* Seem. Hemsl. planted at different sites and treated with different amounts of fertilisers indicates that the type of site affects leaf morphological parameters and fertilisation has a positive effect on leaf parameters and leaf size in the first year after planting.

All measured parameters are significantly higher at the site in Obrenovac (Site 1) than Pambukovica (Site 2), as expected since the soil at Site 1 has better physical properties, contains more humus and has more favourable carbon to nitrogen (C/N) ratio.

The leaves of seedlings treated with a larger amount of fertiliser (240 g per plant) have significantly higher values of all analysed parameters than the leaves of seedlings fed with a smaller amount of fertiliser (120 g per plant) and the leaves of plants in the control area with no treatment. Compared to the leaves from the control area, the plants fed with a smaller amount of fertiliser had significantly higher values of all the parameters of the leaves except for the length of the central nerve, where the difference is not statistically significant.

The analysis of leaf morphological parameters of the investigated species shows that in *Paulownia elongata* S. Y. Hu, the mean values are higher and statistically significant for the properties of leaf area, leaf perimeter, petiole length, the distance between the third and fourth leaf nerves, the number of nerves on the left side of the leaf and the number of nerves on the right side of the leaf. On the other hand, these differences are not statistically significant for leaf lamina length, central nerve length, leaf width, and leaf width at 1 cm from the base.

Based on the obtained results, we can conclude that when introducing these species, we must ensure that the soil is loose and well-aerated. Since this is a fast-growing species, the amount of nutrients added through fertilisation should be adjusted to the physical and chemical properties of the soil so that it contains the right amount. The obtained results point to the potential of these species, and research should be expanded to the subsequent development of the plants until they are harvested in energy plantations where the use of these species can be expected.

Acknowledgements: *This study was carried out within the framework of the Agreement on the implementation and financing of scientific research NIO in 2022, financed by the Ministry of Science, Technological Development and Innovation of the Republic of Serbia, no. 451-03-68/2022-14/200027of 04.02.2022.*

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MORPHOMETRIC CHARACTERISTICS OF *PAULOWNIA ELONGATA* S. Y. HU. AND *PAULOWNIA FORTUNEI* SEEM. HEMSL. LEAVES AND FERTILISATION IN DIFFERENT SITES

Suzana MITROVIĆ, Milorad VESELINOVIĆ, Nevena ČULE, Goran ČEŠLJAR,
Saša EREMIJA, Renata GAGIĆ-SERDAR, Snežana STAJIĆ

Summary

The introduction of new species in the territory of Serbia has become important considering the climatic changes that have become evident in the early decades of the 21st century. These changing environmental conditions have made certain species from different regions of the world suitable for the introduction. Numerous research studies have been conducted to find out whether it is possible and justified to introduce paulownia species. We selected *Paulownia elongata* and *Paulownia fortunei,* species of the *Paulownia* Sieb. & Zucc. genus from the Paulowniaceae family for our research because we already had a plantation established in 1993 near Bela Crkva in Vojvodina to collect seeds for the production of plants that were analysed at other sites. The analysis of the morphological parameters of the leaves of these two Paulownia species was carried out as part of the research dealing with their adaptability to the environmental conditions at two different sites in Serbia. Leaf size and characteristics are important for the process of photosynthesis and the production of nutrients that are directly correlated with the production of the entire plant biomass. Leaves have another important role – acting as filters that clean the air. Based on these facts, a comparative analysis of ten main leaf characteristics was performed (Morphological leaf analysis was carried out following the modified protocol for "Assessments of oak leaf morphology") relative to the soil conditions and different amounts of fertiliser used to feed the plants in the first year after planting. The analysis of the obtained data about the morphological parameters of the leaves of *Paulownia elongata* S. Y. Hu. and *Paulownia fortunei* Seem. Hemsl. planted at different sites and treated with different amounts of fertiliser indicate that the type of site affects leaf morphological parameters and fertilisation has a positive effect on leaf parameters and leaf size in the first year after planting. In both sites and in all treatments, better results were recorded for *Paulownia elongata* S. Y. Hu. The obtained results reveal the potential of these species. Plantations should be established on loose and well-aerated soil. When introducing this species, care should be taken, given that this is a fast-growing species, that the amount of nutrients added matches the chemical properties of the soil so that plants have enough nutrients for growth and development. Research should be continued in the second year of plant development until harvest in energy plantations where these species are expected to be used.

MORFOMETRIJSKE KARAKTERISTIKI LISTOVA VRSTA *PAULOWNIA ELONGATA* S. Y. HU. I *PAULOWNIA FORTUNEI* SEEM. HEMSL. U ODNOSU NA PRIHRANJIVANJE NA RAZLIČITIM STANIŠTIMA

Suzana MITROVIĆ, Milorad VESELINOVIĆ, Nevena ČULE, Goran ČEŠLJAR, Saša EREMIJA, Renata GAGIĆ-SERDAR, Snežana STAJIĆ

Rezime

Introdukcija novih vrsta na području Srbije je od značaja ako imamao u vidu klimatske promene koje su evidentne u prvim dekadama 21 veka. U okviru novonastalih uslova sredine pojedine vrste iz različitih regiona u svetu ukazuju da je njihova introdukcija moguća. U okviru istraživanja koja treba da pokažu da li je introdukcija vrsta paulovnija moguća i opravdana vršena su različita istraživanja. Vrste *Paulownia elongata* i *Paulownia fortunei* roda *Paulownia* Sieb. & Zucc. iz familije Paulowniaceae su odabrane za naše istraživanje jer kod nas već postoji osnovana plantaža u1993. godine kod Bele Crkve u Vojvodini. sa koje je i prikupljeno seme za proizvodnju biljaka koje su analizirane na drugim staništima. Analiza morfoloških parametara listova navedene dve vrste paulovnije vršena je u sklopu istraživanja adaptibilnosti ovih vrsta na uslove sredine dva različita lokaliteta u Srbiji. Veličina i karakteristike lista su od značaja za fotosintetičke procese i produkciju hranljivih materija koje su u direktnoj korelaciji sa produkcijom celokupne biomase biljaka. Značajna je i uloge lista kao filtera za prečišćavanje vazduha. Polazeći od ovih činjence izvršena je komparativna analiza deset osnovnih obeležja lista (Morfološka analiza lista rađena je prema modifikovanom protokolu ”Assessments of oak leaf morphology”) u odnosu na pedološke ulove i prihranjivanja biljaka u prvoj godinirazvoja nakon sadnje sa različitim količinama đubriva. Analiza mernih podataka morfoloških parametara listova vrsta *Paulownia elongata* S. Y. Hu. i *Paulownia fortunei* Seem. Hemsl. posađenih na različitim staništima i tretiranih različitim količinama đubriva ukazuju da tip staništa utiče na morfološke parametre lista i da prihranjivanje pozitivno utiče parametre lista i veličinu listova u prvoj godini nakon sadnje. Na oba lokaliteta i u svim tretmanima bolji rezultati su evidentirani kod vrste *Paulownia elongata* S. Y. Hu. Dobijeni rezultati ukazuju na potencijal ovih vrsta. Plantaže treba formirati na rastresitom i dobro aerisanom zemljištu. Kod introdukcije ove vrste treba voditi računa, obzirom da je ovo brzorastuća vrsta, da se količina hranljivih materija koje se dodaju đubrenjem usklade sa hemijskim osobinama zemlišta kako bi ih bilo u dovoljnoj meri za razvoj i razvitak biljaka. Sa istraživanjima treba nastaviti i u drugoj godini razvoju biljaka sve do žetve u energetskim zasadima gde se može očekivati upotreba ovih vrsta.

CIP - Каталогизација у публикацији
Народна библиотека Србије, Београд

630

SUSTAINABLE Forestry : collection = Održivo šumarstvo = zbornik radova / editor-in-chief Tatjana Ćirković-Mitrović. - 2008, t. 57/58- . - Belgrade: Institute of forestry, 2008- (Beograd : Black and White). - 24 cm

Godišnje. - Je nastavak: Zbornik radova - Institut za šumarstvo = ISSN 0354-1894
ISSN 1821-1046 = Sustainable Forestry
COBISS.SR-ID 157148172