

Assessment of chosen woody plants introduction through growth and physiology characteristics

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Abstract: For the study were used *Vitex agnus-castus* L., *Lagerstroemia indica* L. the both as deciduous shrubs and *Nerium oleander* L. as evergreen one. Plants were planted in two variants, in the ground kept there over year and in pots in winter time removed under plastic house where temperature ranged 3-6 °C. The soil characteristics were comparable in both variants. During of 2011-2013s were assessed processes of adaptability, growth phenophases, year increments and content of chosen metabolites in leaves. Growth phenophases moved on almost one month later at variants in the ground. The year increments at plants planted in pots were significantly less than those planted in the ground and achieved 40-75 cm and 85-95 cm high respectively. The deciduous woody plants planted in the ground were flowered each year, produced germinable seeds and no damage by frost at almost -15 °C. At evergreen *Nerium oleander* all above ground shoots have frozen at -15 °C in winter time and regenerated from root neck and grown over next year. Those a new shoots were survived through next winter at -5 °C and leaves from plants of both planting variants were sampled for analyses of selected metabolites. Results shows almost balanced content of starch from both growing variants and

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achieved 4.73% in pots and 4.34% of dry weight in plants growing in the ground. Total leaf sugar content (in pots/in the ground) achieved 3.72/7.39%, chlorophyll *a* = 0.60/0.79 mg.g⁻¹, chlorophyll *b* = 0.28/0.56 mg.g⁻¹ and carotenoids represents 0.27/0.97 mg.g⁻¹ content in the leaves. The content of analysed metabolites in the leaves of assessed *Nerium oleander* was generally higher at plants in the ground compared with plots and statistically significant. Assessed metabolites should be considered as markers of hardiness in studied woody plants.

Keywords: Mediterranean woody plants, growth, hardiness.

Introduction

Since the earliest times man has developed intentional efforts to be carried plants over great distances for the environmental and life quality improvement. The movement of plant from one country to another and from one region to another that resulted in the expression of new forms and the adaptation to new environment has been critical factor in the development of plants, their selection and breeding for wide form of utilisation by man. Their influence to the natural flora was very active almost 500 year ago but many removed and introduced plants were known and cultivated in ancient civilisations yet before Christmas era (FOSBERG 1959). The main reason for plant movement and introduction include the sources of food (cereals, vegetables, fruits, vine, beverages, drinks, tee plants and coffee, spices and condiments, etc.), ethno medicine and veterinary, timber and biomass production, technical and commercial plants (cotton, crude drugs, packing material) ornamental plants (decoration by leaves, blooms, fruits, bark and crown shape), and also as fuel (SHAH 2005, BIHUŇOVÁ & HREBÍKOVÁ 2010, VEREŠOVÁ & TAKÁČOVÁ 2010, TÓTH, KUCZMAN & FERIANCOVÁ 2016).

Since beginning of civilisation, people have been used plants as medicine and treatment sources throughout human history when between 35thousand and 70thousand plants have been used for medical purposes (FARNSWORTH et al. 1991). Introduction processes passed over different practical ways, theoretical approaches and scientific based decisions.

In the past the first plant introduction forms were realized over market routes predominantly in the course Middle East, western Asia, south Europe, later from Rome Empire territory to the northern Europe (e.g. vine, fruits, and cereals). Other introduction forms were performed by movement of nations, military invasions and discovery of new territories and over see continents as were America (1492), Australia, New Zealand. Important role played in plant introduction from fast Asia (Japan, China, and India) to the Europe through the Dutch East India Company, founded on first half of 18th century. In time of 17th and 18th century many doctors, botanists, gardeners, foresters and agriculturists were undertaken expeditions to the new discovered worlds to be collected plants and placed them to the home botanical gardens, experimental and reproduction bases and nurseries (SVOBODA 1981, BENČAĚ 1982, STEPHEN 1993, SYDNOR 2001, MYERS & BAZELY 2003). On the first half of 20th century a Russian botanist

and geneticist Vavilov elaborated a new scientific approach in introduction processes when described and defined 9 gene centres in terms of the origin and subsequent introduction of the plants into culture with regards in agriculture usage (SALAŠ & LUŽNY 2010).

Other authors define phytogeographical zoning of Earth as so called florozones where potential use of plants for ornamental, horticulture, garden, park and landscape architecture were defined (HENDRYCH 1984). One of most important introduction source is Mediterranean zone from that many plants have been moved to the Central European regions including Slovakia and Czech Republic (SUPUKA 1996). The dominant dendrology object is Arboretum Mlyňany (founded in 1892) where highest collection of evergreen woody plants (in latin-Semper vireo) can be seen originated especially from Mediterranean zone. According to published sources (BENČAĽ 1967, TÁBOR & TOMAŠKO 1992, HOŤKA & BARTA 2012) collection of all evergreen woody plants was inventoried in individual years in follow species and intra specific taxa together: 1925years – 248 taxa, 1952s – 123, 1958s – 126, 1967s – 291, 1992s – 408, 2012s – 273 taxa. While in 1967s has been inventoried 45 taxa of evergreens and deciduous woody species, originated from Mediterranean zone, in 2012s achieved this plant group as far as 157 taxa in living collections of the Arboretum Mlyňany. Mediterranean woody plants was inventoried in another botanical gardens (BG) in current times, in BG – Slovak University of Agriculture in Nitra 62 species (BRINDZA 2002), in BG-MENDELU, Brno (CZ) 123 species and in Lednice Park – MENDELU, Lednice (CZ) 42 species (PEJCHAL & KREJČÍŘÍK 2010).

The all mentioned Botanical gardens and Arboretums does not exceed 250 m above sea level and belongs to the warm climatic zone according to valid state climate classification (MIKLOS & HRNČIAROVÁ et al. 2002). In spite of moderate and warm climate condition in assessed botanical gardens as result of general climate changes brings many disturbances and unbalanced meteorological characteristics over growing seasons. Therefore introduced more sensitive plants need to be sometime protected against frost or irrigated in dry season. Climate geographical conditions of Mediterranean zone are similar, very variable and unbalanced also, but in both regions of Europe it caused warming and desertification processes within last 100 years. As reflection of those climate changes the introduction potential of woody plants from Mediterranean to Central Europe zones has been increased dominantly at ornamental group for park and garden design in settlements. Many contemporary study experiments are aimed for assessing of Mediterranean plants because floristically very reach (ZELENÝ 2012) and for hardiness and drought to be enriched ornamental plant assortment in Central Europe zone. Current state, theoretical and climate assumptions for this intention have described by HOŤKA & BARTA (2012).

Plant introduction and survival process in new above all colder conditions should be assessed at growth and phenological features (SUPUKA 1988, BLUMLER 2005, ALKURDI et al. 2015). Plant response and adaptation to low temperature stress are being very often expressed at the molecular, cell, biochemical and physiological levels also (SANDEVA et al. 2011). According to WEISER (1970) the ability of woody plants to cold resistance is promoted in early autumn in leaves where sugars and

other protective metabolites accumulate, proteins are rearranged the cells become less turgid, growth cessation also occurs. Concerning to drought in the Mediterranean basin vegetative growth during the beginning of summer only happens when roots go deep enough (CASTRO-DIEZ et al. 2005).

Increasing interest on ornamental woody plants of Mediterranean origin is involved also in theory and practice for garden and park design of Slovakian conditions (SUPUKA & FERIANCOVÁ et al. 2008).

Material and methods

The study was conducted during 2011-2014s in Botanical garden of Slovak University of Agriculture in Nitra, Slovak Republic. Three species of Mediterranean evergreen and deciduous shrubs were selected for experiment, *Lagerstroemia indica* L., *Nerium oleander* L., *Vitex agnus-castus* L. The plants were planted on May 3rd, 2011 in two types of plantings each species in triple pieces. Plants planted into the ground and plants planted into pots. The age of planted shrubby individuals was 3 year old and starting average height was as follow (plants in the ground/in the pots): *Lagerstroemia indica* - 126 cm/113 cm, *Nerium oleander* - 81cm/100 cm, *Vitex agnus-castus* - 114 cm/77 cm). Plants planted in the pots have been removed during the winter time from second half of November until end of March inside the plastic house where average temperature were raised from 3°C to 6°C. The plants were irrigated by drip automatic irrigation system during growing season, from spring to autumn. The species was imported from Florence nursery in Italy all the plants were almost in the same size and in the same age, three years old. The phenological observation and assessment was recorded according to methods published by ŠKVARENINOVÁ et al. (2008). The following phenophases and their stages have been assessed, where phenophase rate is designated as follow: 10% - Start, 100% - Full (Tab. 6):

Vegetative phases: leaf bud swelling (LBS 10% - Start, 100% - Full), bud-burst (BB 10%, 100%), leafing (L 10%, 100%), leaf discolouration (LD 10%, 100%), leaf fall (LF 10%, 100%).

Generative phases: flower buds (FB 10%, 100%), flowering (F 10%, 100%), blossom fall (BF 10%, 100%), fruit bearing (FRB 10% - Start, 100% - Full).

Phenological observations were registered in 2012-2013 during all the year round in spring and summer time each two days, and in autumn and winter it was twice during week. Expansion in during whole period of observation we state just in global view on whole work. This expansion was registered with shortening in month (J₁₋₃₁=January 1-31, F- February, M - March, A - April, My - May, Jn - June, Jl - July, Ag - August, S - September, O - October, N - November, D - December) and by number adjusting to the date (e.g. M₁₁ means 11th March).

Results of phenological observation are given in Tab. 6. (Note: vegetative phase LD - leaf discolouration is not presented in table). The meteorological data were acquired from Meteorology and Climate Department of the Slovak University of Agriculture in Nitra as shown in Tab. 1.

Winter hardiness, bio-phenological and reproduction characteristics were evaluated according to BENČAĚ (1967) as shown in Tab. 2, the results are given in Tab. 3.

The total height of plants and year increments were measured every year in the end of growing season by using a meter stick. Measurement of tree height carried out from land level to mean of top branches. Terminal shoot growth (total height and year increment) of 20-25 selected branches was measured on each shrubby plant. Results of total heights are presented in Tab. 4 and year increments in Tab. 6. The nutrient system of soil was assessed through samples taken from the experimental area on December 11th, 2012, from the ground planted plants and plants planted in pots and collected from a depth of 0- 50 mm and 50 - 300 mm as shown in Tab. 7, 8. Nutrients were determined in acceptable form according to Mehlich III (KOBZA 1999). The received values of the soil analysis elements for the experimental area were compared with criteria of soil analysis results according to Mehlich III method and related to critical level contents for agriculture arable land given and approved under Annex No. 5 to Decree No. 338/2005 Coll. Ministry of Agriculture of the Slovak Republic on 6 July 2005 on the procedure for the collection of soil samples (Tab. 8).

For assessment of hardiness at *Nerium oleander* as evergreen woody plant the analyses of chosen metabolites in the leaves were used. The samples were taken in January 2014 when air temperature was - 3 °C at 9 am and during previous night was minimum - 7 °C outside. The pigments (chlorophyll *a*, *b* and carotenoids) were determined according to OLŠOVSKÁ et al. (2013), total sugar and starch contents according to MICHALÍK et al. (1978). Received results are shown in Tab. 9, 10.

An experiment was led out as factorial Randomized Complete Design (RCD) in three replications, the data were analysed with the general linear model procedures in SAS, and Duncan test at level 0.05 was used for the total height plant dates (Tab. 4), year increment (Tab. 5) and biochemical metabolites (Tab. 9, 10) analysed from the plant leaves also (STEEL et al. 1997, OBTULOVÍČ 2010).

Results and discussion

Winter hardiness, bio-phenological and reproduction characteristics

When we have intention to assess of winter hardiness of studied woody plants, we need to introduce climate conditions of the Nitra locality where experiment was established. The climate temperature and rainfalls values are presented in Tab. 1. The marks of winter hardiness were assessed according to methods and their valuation scopes published by BENČAĚ (1967) and presented in Tab. 2. Assessed winter hardiness results of our experiment are given in Tab. 3. The study found that a better hardiness was shown by the deciduous plants. *Lagerstroemia indica* and *Vitex agnus-castus* showed superb winter hardiness, while in *Nerium oleander* all of above ground branches were damaged by winter conditions. *Lagerstroemia indica* and *Vitex agnus-castus* blossomed and gave germinate seeds in each planting type, while *Nerium oleander* planted in pots produced flowers and seeds but not germinated.

Tab. 1. Average temperature in °C and sum of rainfalls in mm (Nitra, 2011-2012- 2013).

Average temperature in °C				Sum of rainfalls in mm			
Month	2011	2012	2013	Month	2011	2012	2013
January	-0.90	1.36	-0.8	January	25	61.1	71.2
February	-0.60	-2.49	1.5	February	6	23.5	75.6
March	5.90	7.41	3.1	March	27	2.8	113.9
April	12.70	11.23	12.1	April	13	36.1	20.4
May	15.80	17.29	15.6	May	48	19.6	77.8
June	19.80	20.86	19.3	June	91	70.1	46.7
July	19.70	22.77	22.8	July	122	61.4	2.1
August	20.90	21.47	21.9	August	152	7.3	73.9
September	17.70	17.02	14.7	September	92	32.7	60.0
October	9.90	10.46	12.1	October	37	76.1	30.5
November	3.00	7.45	6.8	November	1	34.6	71.3
December	2.20	-0.91	2.3	December	42	44.4	11.0
Year Average Temperature	10.51	11.16	11.0	Year sum of Rainfalls	656	469.7	654.4

Tab. 2. Winter hardiness, bio-phenological and reproduction characteristics according to BENČAT(1967).

Winter hardiness (Frost resistance)	Bio-phenological and reproductive characteristics
I, plants do not freeze	VIII, wood species vegetate, but do not bloom
II, 50% of the annual shoots' length freezes	IX, plants bloom, but do not fruit
III, 50-100% of the annual shoots' length freezes	X, they fruit, but give ungerminant seed
IV, older shoots freeze	XI, plants fruit and give germinant seed
V, the aboveground part of the plant freezes to the snow cover height	XII, plants permanently regenerate in a natural way
VI, aboveground part of the plant freezes	
VII, plants are winter-killed	

Tab. 3. Mediterranean species (origin, type of growth, winter hardiness, bio phenological and reproduction characteristics), assessed according to characteristics in both planting types

Species	Origin	Type of growth	Winter hardiness	Phenological and reproduction characteristics	
			rating for plants planted in the ground	Plants in the ground	Plants in pots
<i>Lagerstroemia indica</i> L.	Japan and Southeast Asia	Deciduous	I	XI	XI
<i>Nerium oleander</i> L.	Mediterranean	Evergreen	III -VI	VIII, XII	X
<i>Vitex agnus -castus</i> L.	Mediterranean and central Asia	Deciduous	I	XI	XI

Concerning to woody plants hardiness during which the fall degree of tolerance is achieved requires exposure to period of sub-zero temperature that lead to final phase of plant recovery after winter (Li et al. 2008). The biochemical, physiological and morphological changes associated with low temperature tolerance clearly affect active growth and development and, as a result, a plant must be programmed to recognize and respond to temperatures that are favourable for growth and to the environmental footprints that signal seasonal changes (FOWLER & LIMIN 2004). Wide distributed the red osier dogwood specimen (*Cornus sericea* L.) and its adaptation to a variety of environment conditions, its short generation time and its small stature make it ideal for eco-physiological and genetic studies, that has been used for nearly 40 years in a biotic stress and dormancy research (SVENDSEN et al. 2007).

Woody plant height and the year increment

The total height of plants increased significantly in 2012-2013 for all studied woody plant species comparing with first year 2011 of planting, except of *Nerium oleander* which was planted in the ground (Tab. 4). At this evergreen planted in the ground the height is decreased because all of new shoots growing in spring after damaged branches by foregone winter coldness. Concerning the statistical assessment, the study shows a strong inverse correlation between height of species and planting type in all species as shown in Tab. 4. The highest height of plants showed in *Vitex agnus-castus* -266.7 cm which was planted in the ground in 2013. The ability of woody plants to survive winter is depending on their entry into dormancy state as well as the development of their cold acclimation achieved by a continuous exposure from -5 °C to -15 °C. In late autumn, after leaves have dropped and with the first frosts (up to -3 °C), trees become dormant (Weiser, 1970). Data in Tab. 5 showed that year increment of branches is differing between the species depending in the growth year and planting type. The mean year increment in 2011 of the *Vitex agnus-castus*, which were planted in the ground, achieved 95.36 cm. In 2012 the increment was in *Lagerstroemia indica* - 43.57 cm, *Nerium oleander*- 60.77 cm, *Vitex agnus-castus* - 84.53 cm which were planted in the ground sequentially and the differences were statistically significant with the one to other species and planting types within the same year. The data in 2013 showed the highest mean year increment in *Lagerstroemia indica* achieved by 39.27 cm which was planted in pots. This study observed that the highest mean year increment was in *Vitex agnus-castus* for plants which were planted outside and achieved 95.36 cm in 2011, 84.53 cm in 2012 and 75.04 cm in 2013, while the year increment showed differences with plants which were planted in pots and were as follow: 39 cm in 2011, 58.56 cm in 2012 and 58.69 cm in 2013. Received results were statistically significant. Perhaps due to the limited space in which plants were in pots planted, generally height and average growth increments decrease with increasing age. ASSMANN (1970) stated that the growth rates fluctuate throughout the year according to weather conditions. Environmental conditions both in the previous and current year would affect the annual mean height increment of a tree.

Tab. 4. Total height of plants (in cm) according to species, planting types and year and statistical valuation.

Species	Planting type	Year		
		2011	2012	2013
<i>Lagerstroemia indica</i> L.	In the ground	139.0 ^{e-l}	177.7 ^{c-e}	196.7 ^{bc}
<i>Nerium oleander</i> L.		92.3 ^{o-v}	80.7 ^{q-w}	56.3 ^{vw}
<i>Vitex agnus-castus</i> L.		158.3 ^{c-i}	219.0 ^b	266.7 ^a
<i>Lagerstroemia indica</i> L.	In pots	135.7 ^{e-m}	155.7 ^{c-j}	168.3 ^{c-t}
<i>Nerium oleander</i> L.		130.7 ^{f-n}	158.3 ^{c-i}	114.3 ^{l-t}
<i>Vitex agnus-castus</i> L.		85.7 ^{o-w}	153.3 ^{d-j}	166.7 ^{c-g}

* Means followed with the same letters are not significantly different at 5% level using Duncan test

Tab. 5. Mean and standard deviation of year increment (in cm) for studied species in different planting type within 2011- 2013 years and statistical valuation.

Species	Planting type	2011		2012		2013	
		Mean	SD	Mean	SD	Mean	SD
<i>Lagerstroemia indica</i> L.	In the ground	20.83 ^a	5.73	43.57 ^b	14.40	31.32 ^a	7.00
<i>Nerium oleander</i> L.		34.57 ^a	9.19	60.77 ^b	15.61	36.48 ^a	6.38
<i>Vitex agnus -castus</i> L.		95.36 ^b	32.92	84.53 ^b	20.63	75.04 ^b	17.20
<i>Lagerstroemia indica</i> L.	In pots	20.90 ^a	6.31	21.12 ^a	5.64	39.27 ^b	14.95
<i>Nerium oleander</i> L.		34.57 ^a	8.66	38.57 ^a	11.85	35.86 ^a	6.02
<i>Vitex agnus -castus</i> L.		39.00 ^a	9.70	58.56 ^a	14.69	58.69 ^a	11.20

* Means in the same rows not followed by the same letters are significant at 5% level of probability (Duncan test)

Observation of the growth phenology characteristics

The studied species were in required health generally and some of them have flowered and fruited annually, some plants produced available seeds. The deciduous species which are planted in the ground enter to the growing season late than the plants which were planted in pots and removed to protected area during winter time.

The growth phenophases have presented relatively great differences at the all observed individuals in each year and in both planting types and still greater during all period of observation as shown in Tab. 6. First marks of spring activity that is beginning of leaf bud swelling generally in all species was earlier in plants which were planted in pots and protected during the winter time. In *Lagerstroemia indica* which was planted in pots the leaf bud swelling phenophase was in February while in plants which were planted in the ground were in second half of April in 2012 and in 2013 was in last days of March. *Nerium oleander* which was planted in pots had earlier leaf bud swelling and it was in February comparing with *Nerium oleander* planted in the ground which started in May. *Vitex agnus-castus* in 2012 in plants in pots showed the earliest leaf bud swelling in 20th of January while in 2013 was later but plants in the ground showed the same date for leaf bud swelling phenophase. The beginning

of leaf bud breaking was almost equal in *Lagerstroemia indica*, and *Nerium oleander* which were planted in pots and it was earlier in 2012 comparing with 2013, while *Lagerstroemia indica* which was planted in the ground started in April but in *Nerium oleander* was later in second half of May in 2012 and was later in June during 2013. Leaf bud breaking in *Vitex agnus-castus* started in 31th of January in 2012 and in 2013 was later in 10th of March, while *Vitex agnus-castus* in the ground in both years was in April. The leafing phenophase was first in *Lagerstroemia indica* which was in pots and it started in 3rd of February during 2012, while in 2013 was later in the same plants. *Lagerstroemia indica* in the ground started leafing phenophase in April 2012 and during 2013 was earlier (12 days). In *Nerium oleander* leafing started in 5th March in plants which were in pots and during 2012 was earlier, while *Nerium oleander* in the ground during 2012 was in 23rd March and in 2013 was later in 26th June. *Vitex agnus-castus* in the ground had the same leafing time in both years. *Nerium oleander* planted in the ground during 2013 there were no flowered as shown in Tab. 6 because above ground branches dead by frost during last winter in January 2012 at -15 °C and in spring of 2013 just regenerated a new shoots only but without flowers. *Nerium oleander* and *Vitex agnus-castus* almost started flowering in the same time and the flowering was in July. *Lagerstroemia indica* was the latest species which started flowering in pots 1st August during 2012 and 5th September in 2013 and *Lagerstroemia indica* which was planted in the ground during 2012 flowered in 18th July and in 2013 in 2nd of August. In *Nerium oleander* blossom fall started in 10th August during 2012 and during 2013 was in 26th of July. *Vitex agnus-castus* which planted in pots during 2013 started senescent of flowers earlier than 2012 and in the ground planted plants in both years was the same. *Lagerstroemia indica* in the ground blossom fall started in September and in plants in pots started in October. *Lagerstroemia indica* in pots during 2012 was similar with *Nerium oleander* in 2013. *Vitex agnus-castus* started fruit bearing in August during 2012 in both planting types while in 2013 plants in the ground started earlier in 24th July. Followed phenological phases as were autumn leaves discolouration and defoliation should be seen more particular in Tab. 6. Each species has a genetically determined maximum rate at which it can collect and make use of light, water and nutrients.

Flowering duration in *Lagerstroemia indica* which was planted in the ground during 2012 was 83 days and in 2013 to 71 days. There were differences between *Lagerstroemia indica* in pots and in the ground, when in 2012 achieved 17 days and in 2013 till to 38 days. According to Figs. 1, 2 *Lagerstroemia indica* in pots started in 10% flowering on 214th day of year and 100% blossom fall in 304th day of year and in ground started 10% blossoming in 200th day of year, blossom fall was in 282nd day of year during 2012. Similar results should be seen for 2013 year on Figs. 1, 2. *Nerium oleander* for plants in pots only the duration of flowering was long in 2012 comparing with 2013 (14 days longer). Fig. 2 showed that 10% flowering started in 193rd days of year, 100% blossom fall in 272nd days of year during 2012. In 2013 the 10% flowering started in 188th days of

year, 100% blossom fall in 253rd days of year. *Vitex agnus-castus* in the ground during 2012 had the longest duration of flowering (97 days together) comparing with the same type of planting in 2013 (44 days), also there were difference between plants in pots during 2012 (64 days) and during 2013 (33 days). More particular dates regarding to flowering phenophases are shown on Figs. 1, 2 and Tab. 6.

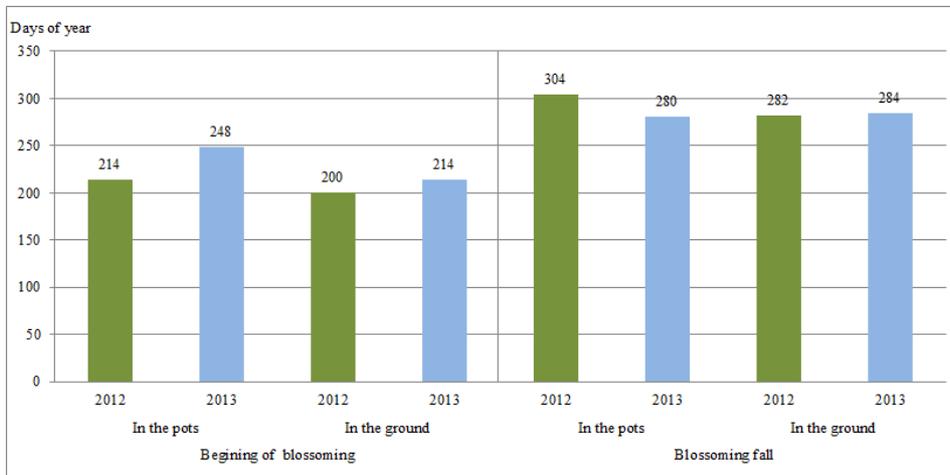


Fig. 1. Beginning and ending of blossoming time at *Lagerstroemia indica* L. planted in pots and in the ground assessed in 2012 and 2013s according to calendar days of year

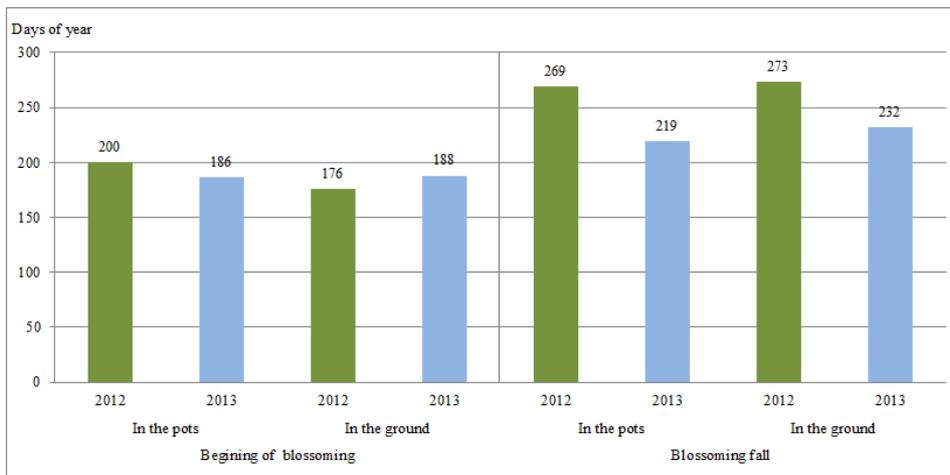


Fig. 2. Beginning and ending of blossoming time at *Vitex agnus-castus* L. planted in pots and in the ground assessed in 2012 and 2013s according to calendar days of year

Tab. 6. Observation of the phenophases in the studied woody plants.

Species	Planting type	Year	Start LBS (10%)	Full LBS (100%)	Start BB (10%)	Full BB (100%)	Start L (10%)	Full L (100%)	Start FB (10%)	Full FB (100%)
<i>Lagerstroemia indica</i> L.	Planting in pots	2012	F ₁₀	F ₂₀	F ₂₃	M ₁	F ₃	F ₂₄	Jl ₂	Jl ₂₆
		2013	F ₂₆	M ₁₀	M ₁₀	M ₂₀	M ₂₇	M ₃₁	Jl ₂₄	Ag ₁₈
	Planting in the ground	2012	A ₁₅	A ₂₀	A ₂₂	A ₂₂	A ₃₀	My ₈	Jl ₃	Jl ₁₂
		2013	M ₂₄	A ₃	A ₈	A ₈	A ₁₈	My ₇	Jl ₁₁	Jl ₂₄
<i>Nerium oleander</i> L.	Planting in pots	2012	F ₁₂	F ₂₃	J ₃₁	M ₃	M ₅	A ₁₂	A ₁₂	My ₈
		2013	F ₂₆	M ₁₀	M ₅	M ₁₅	M ₁₈	A ₁₈	A ₈	A ₂₉
	Planting in the ground	2012	My ₅	My ₁₆	J ₃₁	M ₂₃	M ₂₃	Jl ₁₃	-----	-----
		2013	My ₁₇	My ₂₉	M ₅	Jn ₁₈	Jn ₂₆	Ag ₂	-----	-----
<i>Vitex agnus-castus</i> L.	Planting in pots	2012	J ₂₀	J ₃₁	J ₃₁	F ₂₂	F ₂₈	A ₅	Jn ₈	Jn ₂₉
		2013	F ₂₆	M ₅	M ₅	M ₂₄	M ₂₈	A ₂₅	Jn ₁₈	Jl ₃
	Planting in the ground	2012	A ₈	A ₁₃	J ₃₁	A ₂₇	A ₃₀	My ₂₅	Jn ₁₂	Jn ₂₃
		2013	A ₆	A ₁₄	M ₅	A ₂₈	A ₂₉	My ₂₉	Jn ₁₈	Jn ₂₄
Species	Planting type	Year	Start F	Full F	Start BF	Full BF	Start FRB	Full FRB	Start LF	Full LF
<i>Lagerstroemia indica</i> L.	Planting in pots	2012	Ag ₁	S ₁₀	S ₂₀	O ₃₀	S ₃	S ₂₈	O ₁₅	N ₁₄
		2013	S ₅	S ₂₀	O ₁	O ₇	S ₂₀	O ₇	O ₁₂	O ₂₂
	Planting in the ground	2012	Jl ₁₈	Ag ₁	S ₁₀	O ₈	S ₂₈	O ₈	O ₂₆	N ₁₆
		2013	Ag ₂	Ag ₁₉	S ₁₃	O ₁₁	S ₁₃	O ₉	O ₁₂	O ₂₂
<i>Nerium oleander</i> L.	Planting in pots	2012	Jl ₁₁	Ag ₁	Ag ₁₀	S ₂₈	S ₁₀	N ₇	-----	-----
		2013	Jl ₇	Jl ₂₄	Jl ₂₆	S ₁₀	S ₃	N ₂₀	-----	-----
	Planting in the ground	2012	-----	-----	-----	-----	-----	-----	-----	-----
		2013	-----	-----	-----	-----	-----	-----	-----	-----
<i>Vitex agnus-castus</i> L.	Planting in pots	2012	Jl ₁₈	Jl ₁₇	S ₃	S ₂₅	Ag ₁₇	S ₂₈	O ₂₅	N ₇
		2013	Jl ₅	Jl ₁₅	Jl ₂₄	Ag ₇	Ag ₇	S ₂	O ₂₇	N ₁₂
	Planting in the ground	2012	Jn ₂₄	Jl ₁₁	Jl ₃₀	S ₂₉	Ag ₈	S ₂₈	S ₂₈	N ₂₇
		2013	Jl ₇	Jl ₁₇	Jl ₂₇	Ag ₂₀	Jl ₂₄	Ag ₁₉	S ₁₀	N ₂₀

Explanation: J₁₋₃₁= January 1-31, F- February, M- March, A- April, My- May, J- June, Jl- July, Ag- August, S- September, O- October, N- November, D- December, e.g. M₁₁ means 11th March

Soil properties assessment of the experimental plots from the view point of nutrient regime

Nutrients are essential environmental factors affecting growth and development of plants. The predominant part of the plant nutrients derived from the soil and therefore puts such emphasis on soil nutrient regime. Proper nutrition can be achieved to produce biomass plants in the required quality and quantity so that they can withstand adverse environmental factors. We analyzed the content of elemental nutrients in acceptable form (Ca, Mg, K, P, N and pH),

which are shown in Tab. 7. Received results were compared with limit values defined by criteria under Decree no. 338/2005 Coll. Ministry of Agriculture of the Slovak Republic on 6 July 2005 (Tab. 8). Assess the nutrient regime of soils in comparison with the evaluation criteria for analyzes of soils for arable land and for permanent grassland and their comparison. Compared mutually analysed values (Tab. 7) with limits (Tab. 8) we have found that content of accessible nutrients in both experimental variants (in the soil and in the pots) are in responsible and very high level from point of optimal growing conditions.

Tab. 7. Acceptable nutrient content in soil samples of 2013s according to Mehlich III.

Planting type	Depth (mm)	pH	+Nin (mg.kg ⁻¹)	The nutrient content, mg.kg ⁻¹ (Mehl. III)			
				P	K	Ca	Mg
Ground planted plants	0-50	6.86	13.5	162.5	700	4710	952
Ground planted plants	50-300	6.88	8.6	111.25	537.5	4665	981.5
Pot planted plants	0-50	5.87	27.6	465	900	6700	1369
Pot planted plants	50-300	6.41	25.35	350	712.5	6635	1219

+Nin – Nitrogen inorganic

Tab. 8. Soil analyse characteristics with comparison to content limits defined in Decree No. 338/2005 Coll.

Planting type	Depth (mm)	pH	+Nin (mg.kg ⁻¹)	The nutrient content, mg.kg ⁻¹ (Mehlich III)			
				P	K	Ca	Mg
Ground planted plants	0-50	Neutral	Suitable	Very high	Very high	Very high	Very high
Ground planted plants	50-300	Neutral	Low	Very high	Very high	Very high	Very high
Pot planted plants	0-50	Slightly acid	Good	Very high	Very high	Very high	Very high
Pot planted plants	50-300	Neutral	Good	Very high	Very high	Very high	Very high

+Nin – Nitrogen inorganic

Physiological metabolites content in the leaves

For assessment of adaptation processes and hardiness of studied woody plants were used also methods of selected metabolites content analyses in the leaves of evergreen *Nerium oleander* L., when leaf samples were collected over freeze days in January. The analysed results are presented in Tab. 9, 10. While content of starch was almost balanced between planting types, the total sugar content was distinctly higher in plant leaves planted in the ground of open air with comparison to one planted in pots and protected out of frost as stress factor. The share of sugar content in the levels was statistically significant. The chlorophyll *a* was slightly higher and chlorophyll *b* clearly higher at plantings in the ground with comparison at plantings in pots. Share of *a/b* chlorophyll was higher in plots. Result shows that freeze temperature caused increasing of total sugars and chlorophyll *b* content in leaves of studied woody plant. In the leaves of plants at

the ground plantings has been occurred medium form of necrotic damages by frost. Several studies have shown that environmental factors, especially low temperatures, at high altitudes restrict the physiological processes responsible for tissue formation, such as photosynthesis, respiration, allocation of metabolites and shoot growth (DAY et al. 1989, KOCH et al. 2004). Similar results in changes of chlorophyll characteristics in the leaves of woody species under climate and urban complex stress factor's influences have also been published in other sources (KMEŤ 1999, SUPUKA et al. 2010).

Tab. 9. Dry weight, starch and total sugar contents in leaves of *Nerium oleander* L. influenced by planting typ and statistical valuation. Leaves sampled for analyse on January 23rd, 2014.

Planting type	Studied characteristics		
	Dry weight (mg.g ⁻¹)	Starch (% in dry weight)	Total sugar (% in dry weight)
Planting in the ground	91.85 ^{ef}	4.34 ^{b-e}	7.39 ^c
Planting in pots	91.98 ^{d-f}	4.73 ^{a-c}	3.72 ^d

* Means not followed by the same letters are significant at 5% level of probability

Tab. 10. Chlorophyll a, b, a/b and carotenoids contents in leaves of *Nerium oleander* L. influenced by planting type and statistical valuation. Leaves sampled for analyse on January 23rd, 2014.

Planting type	Studied characteristics			
	Chlorophyll a (mg.g ⁻¹)	Chlorophyll b (mg.g ⁻¹)	Chlorophyll a/b (mg.g ⁻¹)	Carotenoids (mg.g ⁻¹)
Planting in the ground	0.79 ^{bc}	0.56 ^{a-c}	1.41 ^b	0.97 ^{bc}
Planting in pots	0.60 ^{ef}	0.28 ^{b-d}	2.14 ^b	0.27 ^g

* Means not followed by the same letters are significant at 5% level of probability

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