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Tilia spp. – Urban Trees for Future

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Abstract

Tilia spp. (lime trees) as ornamental trees have a privileged place in the European mythology and traditions and this is a reason of their constant presence in the urban landscape. Lime trees are in general very resistant to biotic and abiotic stress, although some sanitary problems may occur (e.g. aphids and other related organisms). *Tilia* spp. are considered a good bioindicator and accumulator of heavy metals. Seasonal accumulations of Pb, Ni, Cr, Mn, Cu and Fe in leaves of *Tilia* spp. in urban and periurban green areas of Western Romania, under different traffic conditions, were analyzed using atomic absorption spectrophotometry. The results confirmed the direct correlation between the traffic intensity and the accumulation of Pb in leaves. By mitigating the effects of air pollution, especially in terms of heavy metals, *Tilia* spp. should be further promoted as urban trees.

Keywords: bioaccumulator, bioindicator, climate change, heavy metals, seasonal accumulation

Introduction

Urban trees play an important role in the urban living space, by bringing social benefits, improving environmental quality (e.g. removing PM10 from urban air) and also contributing to local identity (McDonald *et al.*, 2007; Nowak, 2007). One of the most common urban trees is the Lime tree (*Tilia* spp.), both native species and ornamental varieties. Out of 40 genera and over 350 species of the Tiliaceae family that are to be found especially in the tropical and subtropical regions, in Romania the lime trees are represented by a single genus (*Tilia)* with three species: *Tilia cordata* Mill. (the most common), *T. tomentosa* Moench. (the most drought tolerant), and *T. platyphyllos* Scop. (the most water demanding). Of all trees that tradition invested with special virtues, the lime tree holds a special place both in the Romanian (Eliade, 1995) and in the European imagery (Radouglu *et al.*, 2009) due to its numerous uses and symbols: the sacred tree of Aphrodite in Greek mythology or the sacred tree of the fertility goddess Freia in German mythology (Evseev, 2001), a sacred tree in slavic mythology (Blench and Spriggs, 1999), a national symbol in Slovakia, Slovenia and the Czech Republic (Snoj, 2009). In the Romanian imagery, the lime tree is a constant presence in the traditions related to religious celebrations (Eliade, 1995) and also the most frequently mentioned tree in the poems, songs and stories.

The potential use of *Tilia* spp. as ornamental urban trees and as medicinal plants is well known in all regions of Europe (Ivănescu *et al.*, 1966; Şofletea and Curtu, 2007; Soukand *et al.*, 2013). The air in urban and periurban areas has been polluted with numerous solid and liquid particles in suspension that vary in size, morphology and chemical composition. The *Tilia* spp. leaves have showed significant seasonal accumulation especially for Pb but also for Cr, Fe, Ni, Zn and Mn (Bargagli, 1998; Piczak *et al.*, 2003; Tomaševic *et al.*, 2004b; Anicic *et al.*, 2011). The aim of this

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paper was to provide arguments in order to promote *Tilia* spp. as urban trees by analyzing their social role and their potential as ornamental trees, bioindicators and heavy metal bioaccumulators (Pb).

Materials and methods

Firstly, a literature review has been performed on ornamental role, current health issues, potential use as pollution bioindicator and heavy metal bioaccumulation aspects.

Secondly, the seasonal accumulation of heavy metals in *Tilia* spp. leaves in Western Romania (Timisoara region) has been analyzed. In order to study heavy metal accumulation, samples (minimum 30 g of leaves) have been collected from individuals (11-14 years old) of *Tilia* genus, in street alignments, in locations with different pollution intensity levels: no traffic (Căvăran Forest - *T. tomentosa* and Borlova Forest - *T. platyphyllos*); low traffic (urban area in Timişoara, Albinelor Street - *T. cordata*, *T. tomentosa*, *T. platyphyllos* and rural area Borlova - *T. platyphyllos*, *T. cordata*); medium traffic (urban area Timişoara - I. I. de la Brad Street - *T. cordata*); heavy traffic (road to Timişoara Airport -*T. tomentosa*). Samples were encoded as follows: species (C - *T. cordata*; P - *T. platyphyllos*; T - *T. tomentosa*); locations (AL - Albinelor Street, Timişoara; II - I. I. de la Brad Street, Timişoara; AR - Airport, Timişoara; M - Borlova Forest; B - Borlova village; C - Căvăran Forest); sampling data (M - May; A - August).

The representativeness in both the sampling design and the chemical analyses procedure was ensured. Thus, two sampling operations were performed: in May (average size fresh leaves) and in August (average size mature leaves, before the autumn senescence). Samples were collected during the same week from 10 individuals/location, in three repetitions/individual (90 average size leaves). The leaves were collected from the south-facing upper part of the outer tree canopy and numbered (location, data, species, age). Leaves from the outer canopy accumulate more particles due to the higher transpiration rates (Bargagli, 1998). Random chosen 30 g leaves/individual were washed for 25 seconds with deionized water (pH=5.8-5.9).

The samples were dried at 105 °C for 24 hours. 1-5 g of samples were mixed twice with 5 ml of nitric acid (65%) and the mixture was brought to 80 °C twice (about 2h) until completely dried. 5 ml of de-ionized water was added and the suspension was filtered through special paper filter (180 mm thick, with pores of 20-25 μm). The quantity of de-ionized water was increased up to 50 ml and heavy metal concentrations were measured during the atomic absorption with the spectrophotometer VARIAN 220 FAA. The recordings were performed after the calibration for each microelement, at the following wavelength: Cu 324.7 nm, Fe 259.9 nm, Cr 205.5 nm, Ni 231.6 nm, Pb 220.3 nm.

The variance analysis and the post-hoc Duncan test of multiple comparisons were performed in order to compare the Pb concentrations recorded in the samples for each species, location and time of sampling. All calculations were done using Statistica 10 software.

Results and discussions

Tilia spp. as ornamental trees

Due to their special shape, *Tilia* spp. are used as main elements in designing grandiose and inspiring green areas. The beauty of these species all the year round is underlined by associating *Tilia* spp. with different ornamental shrubs in order to get perspectives focused upon *Tilia* spp. on coniferous background. Combinations in green areas with different species and varieties of *Tilia* spp, which blossom successively, are highly appreciated because of a longer period with flowers and fragrance. Some authors described hybrids (Ivănescu *et al*., 1966; Fricke *et al*., 1980), other authors reported new species (Zare *et al*., 2012) and others considered that genuine inter-specific hybrids are extremely rare (Neville-Jones, 1968). However, molecular markers can be applied to characterize hybridization and species differentiation in *Tilia* genus (Fineschi *et al*., 2003).

Tilia spp. produce a large number of sprouts with larger diameters and heights (Matula *et al*., 2012) and show no decline in sprouting vigour with age (Pigott, 1991). *T. cordata* leaves may represent a reliable indicator for evaluation studies on developmental stability based on the ability of its leaf morphometric characters to buffer their development under contrasting environmental conditions (Velikovic, 2010). The difference in the morphology of the crown as well as in the leaf structure seems to be an advantage for *Tilia* spp. when compared with other popular urban trees in Central Europe, considering the influence on air flow, deposition of air-borne particles, abatement of wind speed, reducing building energy use by lowering temperatures and shading buildings during the summer (Nowak, 2006). Sjöman and Oprea (2010) reported that *T. tomentosa*, *T. cordata* and *T. vulgaris* show long-lasting health and hardiness in street environments of Northern Europe.

In the system of green spaces in Timişoara, the global valuation method of the trees was applied by Ciupa *et al.* (2005), by using three components: ecological value, landscape value (general physiognomy of the species shape, individual size, subjectivity of the observer) and vitality; the results indicated *Tilia* spp. as the most valuable trees in this context. Different authors mentioned the high landscape value of *Tilia* spp. (Ivănescu *et al*., 1966; Neville-Jones, 1968; Jensen, 2003; Sjöman *et al*., 2012; Halajová and Halaj P, 2013). Remarkably old trees as well as the young ones are very common in parks and street alignments in Romania, both in groups and as isolated individuals, mainly in residential areas where citizens decide what species to be planted.

Tilia spp. resistance to biotic and abiotic factors

Climate change seems to increase the frequency of extreme events. In Romania, severe drought (1999-2000, 2003, 2011-2012) alternated with flooding (2005, 2010) and severe frost (2012) with milder winters (2013-2014). *Tilia* spp. seems to be more resistant to drought compared to other native tree species (Simionescu *et al*., 2012). *T. cordata* and *T. platyphyllos* showed sensitivity to backwater (3 months) and to very poor land drainage conditions (Dănescu *et al*., 2011).

Aphids and other related organisms, such as plant lice and mites, are strongly related to *Tilia* spp. in green areas but they only cause minor problems to the lime (Zuparko and Dahlsten 1994).

In flooded areas (2005, 2010, 2014), lime tree collar and roots were relatively fast affected by rot and dieback. *Phytophthora* spp. were among the first organisms to damage the fine roots (Dănescu *et al.*, 2011) and could lead to the death of the stressed plants.

Armillaria spp. proved to be aggressive opportunists only on weakened lime-trees in forests, being followed by *Ganoderma lucidum* and other root destroying fungi. On upper roots and stem of trees in urban areas, the wounded bark of old trees was frequently colonized by *Schizophyllum commune* (sapwood parasite), followed by xylofagus fungi: *Stereum* spp., *Trametes* spp. and *Laetyporus sulphureus* or other Polyporaceae which rot the stem and favour the wind breaks (Chira *et al.*, 2000). Lime-trees were the most resistant tree species to the biotic and abiotic factors in the decade 2001-2010 in Romanian forests (Simionescu *et al.*, 2012), presenting only minor problems (e.g. only 120 acres defoliated by *Caliroa anuplipes* out of a total of infested 742,74 ha).

Tilia spp. as a biomonitor and bioaccumulator

Trees play a special role in reducing the level of fine, "high risk" breathable particles with potential side effects on the environment and on human health (Beckett *et al.*, 2000 from Aničić *et al.*, 2011). Tree leaves have been widely used as an indicator of atmospheric pollution and they are an effective alternative to the usual biomonitoring methods (Braun *et al.*, 2007). *Tilia* spp. is one of the best *indicators* of pollution (especially with chlorine), displaying clear spring (*clorosis*) and summer (*necrosis*) symptoms, in street alignments (Braşov, Romania) and industrial polluted areas (Dej, Romania) (Bolea and Chira, 2005).

Having an adequate morphology, canopy structure and epicuticular wax on the leaf surface, lime-trees have been used as suitable biomonitors of aerial trace elements in leaves in urban areas (*T. tomentosa* - Kovács and Podani, 1986; *T. cordata* - Bargagli, 1998; *Tilia tomentosa, Tilia cordata, Tilia platyphyllos* - Braun *et al.*, 2007). According to Jurani *et al.*, 1993 (from Šomšak *et al.*, 2007), *T. cordata* seems to be a better bioaccumulator of heavy metals than *Acer platanoides*. Different authors (Baycu *et al.*, 2006; Popović *et al.*, 2010; Oroian *et al.,* 2012) reported that of the analyzed urban trees, *A. hippocastanum* accumulated the highest Pb concentrations in leaves and could be used as a monitor of concentrations and seasonal variations of heavy metals in air in urban areas. Recently, the use of plants as bioaccumulators or bioindicators has increased because it makes it possible to forecast air pollution for monitoring purposes.

A low-cost, practical method to assess the quality of the urban habitat consisted of determining the injured leaf area reflectance of *Tilia* spp. out of the leaf surface reflectance (Khavaninzadeh, 2014). The findings of many studies showed that metal accumulation in plants varied, depending on species, tissues and metals but it is obvious that *Tilia* spp. meet many of the requirements of a good bioindicator for heavy metals accumulation.

Heavy metals accumulation in Tilia spp.

There were significant differences ($p<0.05$) between lime-tree species in Cr, Cu, Fe, Ni content and non-significant differences in Pb and Mn content for leaves sampled in May 2014. When analyzing leaves sampled in August, the variance analysis showed significant differences between *Tilia* spp. in heavy metal accumulation except for Fe (p=0.10) and Ni (p=0.07). Our study indicated that only Pb was accumulated by all individuals irrespective of species and locations. The other heavy metals did not show a regular pattern during the growing season (Table 1).

Table 1. Differences in heavy metals content after the seasonal accumulation (May-August, 2014) in *Tilia* spp.

Species*/ Locations**	Heavy metals (mg/kg of dry material)				
	Cr	Cu	Fe	Mn	Ni
C/AL	-0.083	-1.229	14.146	-13.078	-1.822
C/II	0.017	0.031	30.284	-19.291	-1.916
T/AL	-0.302	-2.372	-36.484	-13.667	-2.917
P/AL	-0.22	-1.677	-2.171	-13.019	-3.133
T/AR	-0.01	-1.824	62.075	-22.947	-0.248

*Species = C - *T. cordata*; P - *T. platyphyllos*; T - *T. tomentosa*;
**Urban locations in Timisoara = AL - Albinelor Street; II - Ion Ionescu de la Brad Street; periurban location = AR - Timişoara Airport

Some metals could be used as micronutrients (Fe) or macronutrients (Cu, Zn) but generally, the interpretation of data related to accumulation of heavy metals from the air is difficult (Iordache, 2009). A continuous reduction of annual emissions of Pb has been recorded starting with 2009 in Timisoara. The highest heavy metal emissions were recorded for Pb, Zn and Ni, as coming mainly from fuel combustion (Romania, Timișoara Environment Protection Agency, 2014). The local soil was alkaline, pH > 8.0 (Țenche-Constantinescu, 2011) and this causes lower element solubility and lower availability for plants (Tomaševic *et al.*, 2004a; Serbula *et al.*, 2013). Pb solubility in soils with various conditions was very low (Adriano, 1986; Alloway, 1995) and terrestrial plants accumulated less Pb from soil than from the atmosphere through stomata and the accumulated quantity depended on air pollution, particle size and climate. Under such environmental conditions, the atmospheric origin of the elements accumulated in leaves may be assumed: *Achillea millefolium* and *Hordeum vulgare* (Pilegaard and Johnsen 1984), *Quercus ilex* (Alfani *et al.*, 1996), *Populus* spp. (Gregger, 1999), *Salix* spp. (Iordache, 2009), *Picea abies* (Hovmand *et al.*, 2009) and *Tilia* spp. (Serbula *et al.*, 2013).

There were highly significant differences in Pb accumulation ($p<0.001$) by comparing: all species under similar traffic conditions at the end of the growing season (C/AL/A; P/AL/A; T/AL/A); different species or individuals of the same species, under different traffic conditions and in the same sampling period and different species or individuals of the same species, under the same traffic conditions and at different moments of sampling (seasonal accumulation) (Table 2). Moreover, we found significant differences in Pb accumulation ($p<0.05$) by comparing *T. platyphyllos* and *T. cordata* under different traffic conditions at the end of the growing season (P/B/A; C/II/A). However, no significant differences in Pb accumulation ($p>0.05$) were found when comparing species at the beginning of the growing season under different traffic conditions (C/AL/M; C/II/M; P/AL/M; T/AL/M; T/AR/M*)*. The Pb accumulation in leaves at the beginning of the growing season was very low for all species and locations (minimum 0.01280 mg/kg Pb - T/AR/M and maximum 0.3631 mg/kg Pb - T/AL/M).

There were highly significant differences in Pb accumulation among all species of lime-tree studied under similar traffic conditions (Albinelor Street, Timisoara) at the end of the growing season. The differences between *T. cordata* and *T. tomentosa*, between *T. platyphyllos* and *T. tomentosa* and also among individuals of the same species in different traffic conditions in the same sampling period, were also highly significant. The lowest average accumulation during the growing season was recorded in the area with low traffic for *T. tomentosa* (7.03 mg/kg Pb - T/AL) and the highest average accumulation was recorded in the area with the heaviest traffic for *T. tomentosa* (17.83 mg/kg Pb - T/AR). Similar results were reported in various deciduous broadleaves: *Quercus ilex* (Alfani *et al.*, 1996), *Tilia* spp. and *Aesculus hippocastanum* (Sućur *et al.,* 2010), *Acer platanoides, Aesculus hippocastanum* and *Betula pendula* (Petrova *et al.,* 2014), in which Pb was mainly accumulated due to the traffic intensity in the sampling location. Recent studies indicated that Pb has exceeded the maximum admitted level in Cluj-Napoca, Romania, under different traffic conditions, in the leaves of *Acer platanoides, A. pseudoplatanus, Aesculus hippocastanum, Betula pendula, Juglans regia, Picea pungens var. glauca, Picea abies, Pinus nigra, Tilia cordata, Robinia pseudoacacia, Thuja occidentalis* (Viman *et al.*, 2011).

There were highly significant differences in Pb seasonal accumulation not only among all species of lime-tree studied but also among individuals within species, in the same traffic conditions and different sampling periods (seasonal accumulation). The Pb accumulation at the end of the growing season is high for all species and locations (minimum 7.4 mg/kg Pb - T/AL/A and maximum 17.84 mg/kg Pb -T/AR/A). The highly significant differences in Pb accumulation among the lime-tree species studied at the end of the growing season may be in relation with leaf shape and size (Hovmand *et al.*, 2000; Alfani *et al.*, 1996). Seasonal accumulation of other potential toxic metal, arsenic (As), in the leaves of *Tilia* spp. and *Aesculus hippocastanum* was reported in the Belgrade urban area (Sućur *et al.*, 2010). However, As and Pb do not have any physiological function in plants and could be toxic (Adriano, 1986; Madejón *et al.*, 2004). The seasonal accumulation of Pb may be explained as a detoxification process by passive sequestration of toxic metals in senescing foliage (Aznar *et al.*, 2009 from Popovic *et al.*, 2010). Significant seasonal accumulations of heavy metals in the leaves of *Tilia* spp. were reported (Bargagli, 1998; Piczak *et al.*, 2003; Tomaševic *et al.*, 2004b; Anicic *et al.*, 2011).

Table 2. Multiple comparisons of the average Pb content for *Tilia* spp. leaves at the beginning and at the end of growing season, in areas with different pollution level from Western Romania

Dataset*/ Pb average content (mg/kg of dry material)	C/AL/M 0.06850	C/AL/A 15.36400	C/II/M 0.27650	C/II/A 15.95800	C/B/A 13.72000	P/AL/M 0.17130	P/AL/A 10.45400	P/M/A 15.08000	P/B/A 14.68600	T/AL/M 0.36310	T/AL/A 7.4000	T/AR/M 0.01280	T/AR/A 17.84300
C/AL/M													
C/AL/A	**0,000017**												
C/II/M	0.699553	**0.000016**											
C/II/A	**0.000011**	0.239103	**0.000017**										
C/B/A	**0.000017**	**0.002576**	**0.000024**	**0.000074**									
P/AL/M	0.838130	**0.000017**	0.834406	**0.000017**	**0.000021**								
P/AL/A	**0.000024**	**0.000024**	**0.000046**	**0.000021**	**0.000052**	**0.000029**							
P/M/A	**0.000017**	0.572612	**0.000017**	0.100915	***0.010700***	**0.000016**	**0.000029**						
P/B/A	**0.000016**	0.206397	**0.000021**	***0.020511***	0.056919	**0.000017**	**0.000046**	0.434093					
T/AL/M	0.599614	**0.000017**	0.863366	**0.000016**	**0.000029**	0.722013	**0.000052**	**0.000021**	**0.000024**				
T/AL/A	**0.000029**	**0.000021**	**0.000052**	**0.000017**	**0.000046**	**0.000046**	**0.000108**	**0.000024**	**0.000029**	**0.000108**			
T/AR/M	0.911895	**0.000011**	0.638529	**0.000011**	**0.000016**	0.768870	**0.000021**	**0.000017**	**0.000017**	0.543310	**0.000024**		
T/AR/A	**0.000011**	**0.000055**	**0.000017**	**0.000380**	**0.000024**	**0.000011**	**0.000017**	**0.000046**	**0.000029**	**0.000017**	**0.000016**	**0.000011**	
T/C/A	**0.000021**	**0.000042**	**0.000029**	**0.000024**	0.140051	**0.000024**	**0.000110**	**0.000147**	**0.001351**	**0.000046**	**0.000052**	**0.000017**	**0.000021**

(Duncan test, bold marked effects are highly significant at $p < 0.0001$; italic marked effects are significant at $p < 0.05$)
*Species (C - *T. cordata*; P - *T. platyphyllos*; T - *T. tomentosa*); locations (AL - Albinelor Street, Timişoara; II - I. I. de la Brad Street, Timişoara; AR - Airport, Timişoara; M - Borlova Forest; B - Borlova village; C - Căvăran Forest); sampling data (M - May; A - August)

Conclusion

Lime tree species appeared to be highly resistant to abiotic and biotic stress. No major pest or disease of *Tilia* spp. was reported in the recent period in Western Romania. Our results indicated that the Pb content significantly increased during the growing season from very low values in May to much higher values in August in *Tilia* spp. leaves for all individuals studied and locations. The highest Pb content values were recorded for *T. tomentosa* in the area with the heaviest traffic and towards the end of the growing season. This supports the hypothesis of a self-detoxification mechanism of *Tilia* spp. by leaves fall. Other heavy metals analyzed (Cr, Cu, Fe, Mn, Ni) did not have regular accumulation in leaves during the growing season, probably due to different metabolic aspects. In spite of the increasing tendency to introduce exotic tree species in green areas, in the context of climate change, *Tilia* spp. are highly recommended to be used in urban landscapes. These tree species showed a high potential to improve the urban environment and also have an important ornamental, ecological and socio-economic value.

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