PHOTOSYNTHETIC CAPACITY IN *AMORPHA FRUTICOSA*, *ACER NEGUNDO*, *AILANTHUS ALTISSIMA* AND *ELEAGNUS ANGUSTIFOLIA*, THE INVASIVE PLANTS VS. NATIVE PLANT IN DANUBE DELTA BIOSPHERE RESERVE AREAS

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Abstract

The purpose of this study is to analyse gas- exchange parameters and chlorophylls synthesis in order to establish the photosynthetic capacity of some ligneous plants with invasive behaviour in comparison with native trees in areas of Danube Delta. The investigation was carried nearly of Razelm Lake (Beştepe hill), a xerophilous coastal meadow with clumps of trees (fallow vine plantation) and in three other plots (Plaur I, Plaur II and Pătlăgeanca) situated along Chilia branch. Photosynthesis rate registered lower values in *Amorpha fruticosa* (3.53 µmol m^{-2}-s^{-1}) in comparison with *Populus alba*, young plantation (33 µmol m^{-2}-s^{-1}), lower value in *Ailanthus altissima* (13.5 µmol m^{-2}-s^{-1}) in comparison with *Fraxinus ornus* (14.54 µmol m^{-2}-s^{-1}), comparable values in *Acer negundo* with *Amorpha fruticosa* (close to 3.5 µmol m^{-2}-s^{-1}). Generally, the investigated invasive plant registered a lower respiration rate than non-invasive/native species. Transpiration was direct proportional with photosynthesis rate and with stomatal conductance. Photo-assimilatory pigments represented by chlorophyll a, chlorophyll b and carotenoids as total registered in *Ailanthus altissima* being by approximative 1.4 times higher in comparison with *Crataegus monogyna* and *Fraxinus ornus* (Beştepe station), by 1.5 times higher in *Amorpha fruticosa* than *Acer negundo* in ruderal area (Plaur I) and having close values at *Amorpha fruticosa* and *Populus alba* in riparian plantation (Plaur II). Analysis of photosynthetic capacity revealed the competition strategy between invasive plant and native or even among invasive, especially co-dominant species such as *Amorpha fruticosa* vs. *Populus alba*, *Amorpha fruticosa* vs. *Acer negundo*, *Ailanthus altissima* vs. *Fraxinus ornus and Eleagnus angustifolia* against *Salix alba* in studied areas.

Key words: *Amorpha fruticosa, Acer negundo, Ailanthus altissima*, *Eleagnus angustifolia*, invasive, gas-exchange parameters

Invasive plants species appear mostly in different disturbed environments, ecosystems characterized by higher availability of resources, sometimes affected by climatic extremes. Species adapted to low-resource system show traits associated with its resource conservation, such as slow growth, high tissue longevity, and resource efficiency (Funk J.L., 2013). Last decades dryness and drought have the greatest intensity and frequency in Romania. Danube Delta annual average of precipitation decrease to the sea shoreline and the average temperature increases, natural habitats of Natura 2000 being exposed to the spreading potential of invasive species.

Amorpha fruticosa (false indigo bush) is one of the most important invasive terrestrial plant species in Romania with other species such as *Ailanthus altissima*, *Acer negundo,* and *Fraxinus pennsylvanica*. In the last 20 years, in Romania within framework of Kyoto Protocol, Afforestation of Degraded Agricultural Land Project, 6500 ha in Romania was destined to mitigate climate change to reduce CO_2 emissions by planting different ligneous species. Within the located plots, *Amorpha fruticosa* was introduced in the riparian areas of the Danube, including Danube Delta, thus this species invaded the gaps between the trees and even the young white poplar plantations (Ciuvăț A. L., 2016; Doroftei M., Covaliov S., 2009). It has a negative impact on native wetland ecosystems and control measures have been applied exclusively in protected areas. *Eleagnus angustifolia* was introduced in forest cultures from the coastline cordon area and on sand banks in order to fix sand, being present on a large surface. Generally, it appears in the fluvio-maritime delta, in all the localities along the Danube Delta as well as in the fluvial Delta. Nowadays, it is considered subspontaneous species and due to spreading potential, it competes and eliminates *Hippophaë* sp. *Ailanthus altissima*, cultivated as ornamental, also could be considered a subspontaneous species, being present around

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localities and in forest plantations. In last years, these species are catalogued as invasive (Doroftei M., Anastasiu P., 2014; Doroftei M., Covaliov S., 2009).

The purpose of this study is to analyse foliar gas- exchange parameters, chlorophyll synthesis and parameters of water status in order to establish the photosynthetic capacity of some ligneous plants with invasive behaviour in comparison with native or non-invasive species in areas of Danube Delta Biosphere Reserve.

MATERIAL AND METHOD

Study sites

Four natural and anthropic site from Danube Delta Biosphere Reserve were selected from the geographical area:

1. Pătlăgeanca (45°13′45″N 28°44′45″E) a riparian area with *Salix alba* and *Eleagnus angustifolia* along of Chilia brunch on aluvisols.

2. Plaur I (45°19′26″N 28°49′48″E), a ruderal area near dyke with *Amorpha fruticosa* and *Acer negundo* as co-dominant species. Aluviosol is the soil type.

3. Plaur II (45°16′31.4″N 29°39′21.8″E) is the riparian coppice with young plantation of *Populus alba* (5 - 10 years-old). In the vicinity to the old plantation (*Populus x canadensis*) there are dense bushes of *Amorpha fruticosa*. Type soil is aluviosol gleic eutric (with relatively high fertility).

4. Beștepe hill (45°05′32″N 29°00′53″E), a fallow vine plantation with *Ailanthus altissima, Crataegus monogyna* and *Fraxinus ornus* as resentative tree species beside a xerophilous grassland and shrubs on a slope dominated by limestone-rocks, with calcareous soil or different type of chernozem.

Gas-exchange parameters, *in situ* determination of photosynthesis rate (A), respiration rate (R), transpiration (E), respiration (R) and also, instantaneous water-use efficiency (WUE=A/E ratio) were analysed with Lci photosynthesis portable system (ADC BioScientific, UK).

The spectrophotometric determination of photo-assimilating pigments was by fresh leaves solvation in 85% acetone (Meyer-Berthrand modified by Ştirban, 1985). The results were expressed in mg/g fr.w. as fresh weight.

Dry matter of plant leaves was analyzed by heating at 105℃ for 2 hours until constant weight through gravimetric moisture method.

RESULTS AND DISCUSSIONS

Analysis of gas-exchange parameters *in situ* showed that photosynthesis rate registered a higher value of 19.94 µmol m^{-2} s^{-1} in *Eleagnus angustifolia* in comparison with 18.41 µmol m^{-2} s^{-1} registered in *Salix alba*, a native tree from Pătlăgeanca plot (*table 1*). An interesting aspect is observed in Plaur I where photosynthesis rate, transpiration and stomatal conductance have close value in the two invasive species, *Amorpha fruticosa* and *Acer negundo*. Thus, photosynthesis rate was almost 3.4 µmol m^{-2} s^{-1} in both species, transpiration was 1.18 in *Amorpha fruticosa* and higher by 1.52 mmol m^{-2} s^{-1} in *Acer negundo*, leading to a water use efficiency (A/E) with a higher value in *Amorpha fruticosa* of 3.033 in comparison with 2.24 in *Acer negundo*. In that case, in Plaur I plot, *Amorpha fruticosa* registered a respiration rate of 0.41 µmol m^{-2} s^{-1} being by 4.4 times lower than that 1.815 µmol m^{-2} s^{-1} registered in *Acer negundo*. In Plaur II, a riparian coppice, young *Populus alba* tree registered highest photosynthesis rate of 33.74 µmol m^{-2} s^{-1}, highest transpiration rate of 10.22 mmol m^{-2} s^{-1} and also highest stomatal conductivity of 2.19 mmol m^{-2} s^{-1} than *Amorpha fruticosa* from the same plot and also of all investigated species (*table 1*). Water use efficiency was slightly higher in *Populus alba* with the value of 3.46 in comparison with 3.20 obtained in *Amorpha fruticosa.* Although *Populus alba* is a water-consuming tree and in this case a young one (5-10 years-old), recorded respiration was 6.75 times higher in *Amorpha fruticosa* (4.05 µmol m^{-2} s^{-1}) than in *Populus alba* (0.6 µmol m^{-2} s^{-1}). In Beștepe hill, *Fraxinus ornus* registered higher value of photosynthesis rate of 14.54 µmol m^{-2} s^{-1}, even of transpiration by 5.74 mmol m^{-2} s^{-1} and stomatal conductance by 0.26 mmol m^{-2} s^{-1} than obtained in *Ailanthus altissima. Ailanthus altissima* registered increased water use efficiency with a value of 3.93 in comparison with 2.69 obtained at *Fraxinus ornus.* Respiration in *Fraxinus ornus* registered an increased value by 7.96 µmol m^{-2} s^{-1} being 2.6 times higher than that showed by *Ailanthus altissima (table 1).* Internal concentration of CO_2 and stomatal conductance *gs* registered lower values in invasive plant in comparison with its ligneous cohabitant. Another characteristic for invasive plant is a lower respiration rate in comparison with other cohabitants. Thus, *Acer negundo* registered a respiration with almost 2 times higher (1.815 µmol m^{-2} s^{-1}) than *Amorpha fruticosa* (0.41 µmol m^{-2} s^{-1}) in ruderal area at Plaur I. Although, an exception occurred such as in case of *Eleagnus angustifolia*, an invasive, subspontanous species which registered a respiration by 2.6 times higher (3.63 µmol m^{-2} s^{-1}) than *Salix alba* (1.38 µmol m^{-2} s^{-1}), a native species. Subsequently, water use efficiency in *Salix alba* obtained a higher value of 7.70 than that showed by *Eleagnus angustifolia* of 6.83 (*table 1*).

Table 1

Photosynthetic gas-exchange parameters in representative invasive and native plants at studied sites

Station	Species	Ci (µmol mol^{-1})	A (µmol m^{-2} s^{-1})	E (mmol m^{-2} s^{-1})	gs (mmol m^{-2} s^{-1})	R (µmol m^{-2} s^{-1})	Wue (µml/mmol)
Pătlăgeanca	*Eleagnus angustifolia*	319 ± 23	19.94 ± 3.38	3.09 ±0.16	1.97± 0.27	3.63 ±0.87	6.83 ± 1.53
	Salix alba	222 ± 19.45	18.41 ±1.04	2.65 ± 0.27	0.24 ± 0.04	1.38 ± 0.5	7.70 ± 0.74
Plaur I	*Amorpha fruticosa*	275 ± 15.31	3.36 ± 0.30	1.18 ± 0.06	0.04 ± 0.004	0.41 ± 0.11	3.03 ±0.31
	Acer negundo	323± 11.15	3.47 ± 0.42	1.52± 0.03	0.05 ± 0.0	1.815 ± 0.34	2.24 ±0.25
Plaur II	*Amorpha fruticosa*	262 ± 15.78	3.97 ± 0.32	1.47± 0.18	0.042 ± 0.0	4.05 ± 0.73	3.20± 0.14
	Populus alba	287 ± 11.18	33.74 ± 1.98	10.22 ± 0.97	2.19 ± 0.81	0.6 ± 0.44	3.46 ± 0.29
Beștepe	*Ailanthus altissima*	262 ± 10.76	13.9 ± 1.27	3.53 ± 0.16	0.22 ± 0.01	2.52 ± 0.76	3.93 ± 0.35
	Fraxinus ornus	254± 16.8	14.54 ± 0.93	5.74 ± 0.38	0.26 ± 0.02	7.97 ± 0.63	2.69 ± 0.02

Legend: Ci - Substomatal cavity CO_2 concentration, A - photosynthesis rate, R - respiration rate, E - transpiration rate, gs - stomatal conductance, Wue - water use efficiency, Mean ± standard error

Analysis of photosynthetic pigments showed the invasive plant registered close value with native plant, or even with other representative spontaneous plant. In invasive plant are observed that chlorophyll a have a slightly higher value, such as in *Amorpha fruticosa* with 2.56 mg/g fr. w. in comparison with *Acer negundo* with a value of 1.86 mg/g fr. w. at Plaur I, also, *Ailathus altissima*, invasive species registered 1.23 mg/g fr. w. in comparison with 0.796 mg/g fr. w. in *Fraxinus ornus* and respectively, by 0.93 mg/g fr. w. in *Crataegus monogyna*, non-invasive in Beștepe hill plot (*figure 1*).

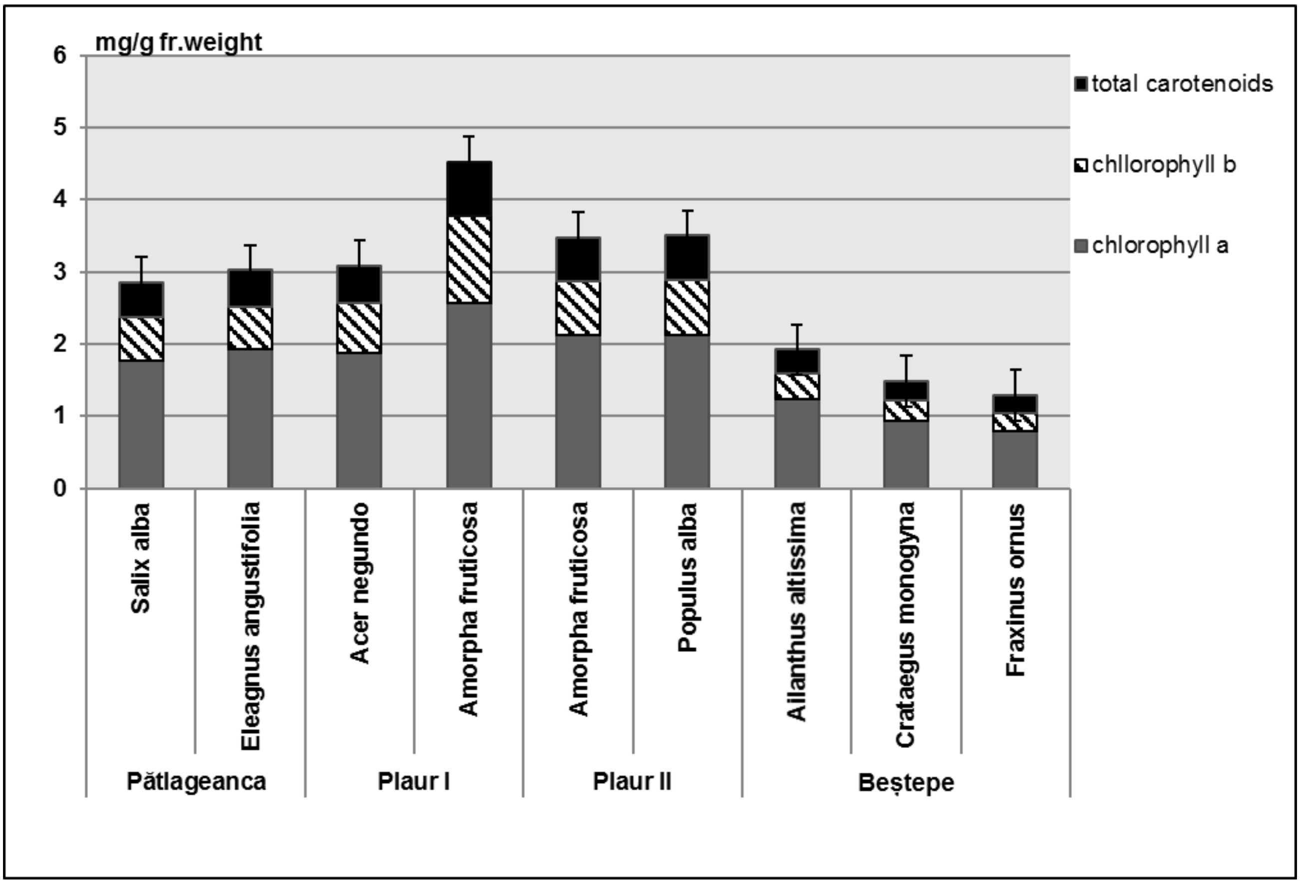

Figure 1 **Graphic representation of chlorophyll a, chlorophyll b and total carotenoids in invasive and native ligneous plant in investigated sites**

Eleagnus angustifolia with 1.93 mg/g fr. w., showed a higher value than 1.77 mg/g fr. w. registered in *Salix alba,* native species at Pătlăgeanca plot. In Plaur II, *Amorpha fruticosa* and *Populus alba* registered very close values of chlorophyll a, approximatively of 2.12 mg/g fr. w., also chlorophyll b being of 0.7 mg/g fr. weight and total carotenoids with 0.61 mg/g fr. Total carotenoids obtained higher values by 0.60 mg/g fr. w in *Salix alba*, a native species than 0.57 mg/g fr. w obtained in *Eleagnus angustifolia,* invasive species at riverside cordon area from Pătlăgeanca. *Amorpha fruticosa* showed a value of 0.74 mg/g fr. higher than *Acer negundo* which obtained 0.61 mg/g fr. in Plaur I plot and also higher value in *Ailathus altissima* 0.34 mg/g fr. than those registered in *Crataegus monogyna* by 0.26 mg/g fr. and *Fraxinus ornus* by 0.25 mg/g fr. (*figure 1*). Generally, photo-assimilatory pigments represented by chlorophyll a, chlorophyll b and carotenoids as total registered higher values in *Ailanthus altissima* (1.92 mg/g fr.w) than in *Crataegus monogyna* (1.28 mg/g fr.w) and also than *Fraxinus ornus* (1.48 mg/g fr.w) in Bestepe station, being higher in *Amorpha fruticosa* (4.52 mg/g fr.w) than *Acer negundo* (3.08 mg/g fr.w) in ruderal area (Plaur I) and close value in *Amorpha fruticosa* and *Populus alba* (close 3.5 mg/g fr.w) in riparian plantation (Plaur II) (*figure 1*).

Some invasive species such as *Alnus formosana* had a higher plasticity in photosynthetic pigments than the native species at different irradiance, which contribute to successfully invasion in high-irradiance locations through better photoprotection (Liu S. et al, 2016). In present study, the chlorophyll a and total carotenoids registered values from one to around 1.5 three times higher than in non-invasive or native plant. *Amorpha fruticosa* developed a competitive strategy perfectly adjusted to the other ligneous plant in investigated plots, such as against *Acer negundo* (representative, other invasive plant) and against *Populus alba* (native young tree) (*figure 1*).

Table 2

Chlorophylls ratio (a/b) and chlorophylls/total carotenoids (a+b/c) ratio in invasive and native/non-invasive species in studied sites

Station	Species	Total pigments (a+b+c)	a/b	(a+b)/c
Pătlageanca	*Salix alba*	2.863± 0.05	2.931 ± 0.14	4.938 ± 0.093
	Eleagnus angustifolia	3.025 ± 0.042	3.336 ± 0.132	4.916 ± 0.1
Plaur I	*Acer negundo*	3.086 ± 0.035	2.71 ± 0.14	4.93 ± 0.1
	Amorpha fruticosa	4.523 ± 0.05	2.106 ± .139	5.064 ± 0.18
Plaur II	*Amorpha fruticosa*	3.478 ± 0.052	2.859 ± 0.16	4.681 ± 0.2
	Populus alba	3.51 ± 0.03	2.745 ± 0. 16	4.714 ± 0.21
Beștepe	*Ailanthus altissima*	1.923 ± 0.046	3.465 ± 0.147	4.733 ± 0.15
	Crataegus monogyna	1.487 ±0.05	3.2230 ± 0.12	4.588 ± 0.16
	Fraxinus ornus	1.29 ± 0.048	3.309 ± 0.15	4.116 ± 0.14

Photooxidative protection of invasive plant is increased, chlorophylls a+b / total carotenoids ratio are higher having values from 1 to 1.15 times higher in *Amorpha fruticosa* vs. *Acer negundo* and respectivelly, in *Ailanthus altissima* vs. *Fraxinus ornus.* In riparian plantated corridors, *Salix alba* vs. *Eleagnus angustifolia* and respectively *Populus alba* vs. *Amorpha fruticosa* realized close values at chlorophylls ratio as well as in chlorophylls (a+b)/total carotenoids (*table 2).* The exotic tree species *Ailanthus altissima* is shade intolerant but succeeds in low-light area because it requires only a small place to rapidly growth and this species are able to take advantage of high-light conditions and grow rapidly in response to natural or human-produced canopy gaps (Funk J., 2013).

Water content analysis showed that leaves of invasive plant registered a higher value than non-invasive species but smaller than native species (*figure 2*).

Thus, *Salix alba* showed a higher value of 39.83 % in comparison *Eleagnus angustifolia* with 32.82 % in Pătlăgeanca. In Plaur I, *Amorpha fruticosa* with 35.84 % retained a more water in leaves having almost by 1.3 times more humidity than *Acer negundo* which registered 27.74 %.

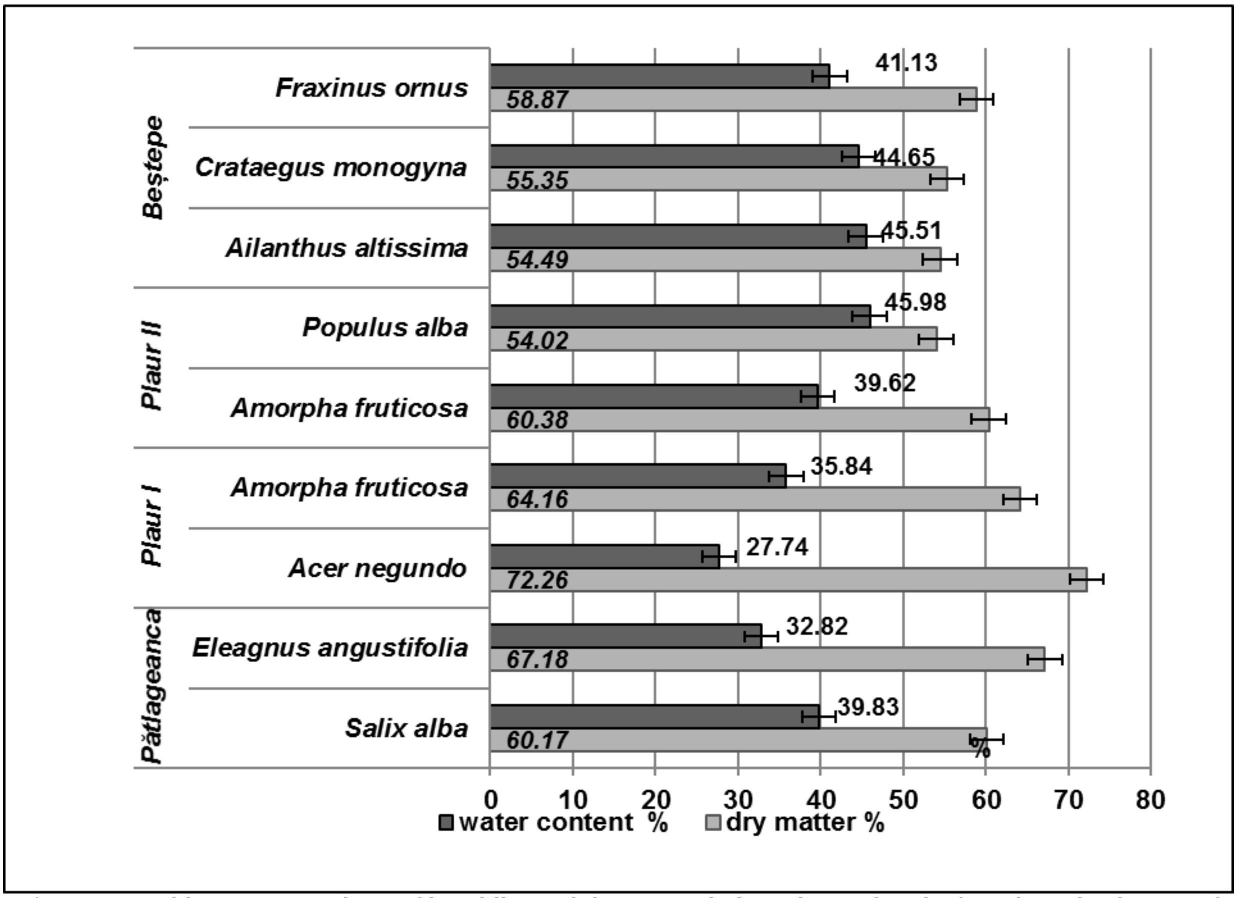

Figure 2 **Graphic representations of humidity and dry matter in invasive and native/non-invasive leaves of ligneous plant in investigated sites**

In riparian young plantation, *Populus alba*, native, young tree, accumulated a water value of almost 50 % being that 1.2 times higher than *Amorpha fruticosa* with a value of 39.62 % (Plaur II).

In Beștepe hill, at fallow land, *Ailanthus altissima* registered close value of water content, with *Crataegus monogyna*, which was approximately 45 %, but higher than *Fraxinus ornus* which accumulated 41 % water in their leaves.

Generally, water status is linked with a strategy to adjust and modulated the plant resistance over temperature variation. Invasive plant it was observed to have mechanism at leaf level to reduce the water loss and thus humidity maintain during vegetation stage (Acatrinei L., 2016; Cavaleri M. A., Sack L., 2010).

CONCLUSIONS

Gas-exchange parameters at invasive ligneous species analysed from different plots of Danube Delta Biosphere Reserve showed an increased photosynthesis rate combined with a lower internal CO_2 and a lower stomatal conductance. These ligneous invasive realized a higher water use efficiency among investigated plant with the exceptions of *Populus alba* and *Salix alba*, both native species. Also, there are observed a lower respiration rate in invasive than non-invasive/native, with the exception of *Populus alba* young tree, (young plantations) which registered a lower value in comparison with *Amorpha fruticosa* .and respectively, *Salix alba* in comparison with *Eleagnus angustifolia* in the riparian corridors. Generally, the concurrence strategy was observed between invasive vs. invasive, such as *Amorpha fruticosa* against to *Acer negundo* or invasive vs. native, between *Eleagnus angustifolia* and *Salix alba* and respectively, *Amorpha fruticosa* against *Populus alba.* This photosynthetic strategy was revealed by close value or even higher value of photosynthesis rate, contents of photo-assimilatory pigments (especially, chlorophyll a and carotenoids pigments) or chlorophylls/carotenoids ratio observed in invasive ligneous species.

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