INTERSPECIFIC VARIABILITY OF SOME ACCESSIONS OF *SALIX* SP. COLLECTED FROM OLTENIA REGION, ROMANIA

***Mihaela CORNEANU*[1], *Cornelia BUZATU-GOANTA (nee HERNEA)* [1], *Wilhelm HOLLERBACH*[2], *Adrian HORVAT*[3]**

(1) University of Life Sciences "King Michael I" from Timisoara, Romania, Faculty of Engineering and Applied Technologies, 109 CaleaAradului, Timisoara; Romania
mihaela.corneanu@usab-tm.ro, cornelia.buzatu-goanta@usab-tm.ro
(2) University of Life Sciences "King Michael I" from Timisoara, Romania, Faculty of Agriculture, 109 CaleaAradului, Timisoara; Romania
wilhelm.hollerbach@rebina.ro
(3) University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, 3-5 CaleaManastur, Cluj-Napoca, Romania
adrian.horvath@rebina.ro
Corresponding author email: cornelia.buzatu-goanta@usab-tm.ro

Abstract
The Salix genus includes many species with various uses: wood production, biomass, animal feed, source of salicylic acid, land reclamation, biofuel, a.o. In order to establish a collection of potential genitors, source of resistance genes to various stressors, 39 accessions were collected in 2015 from Banat and Oltenia, Romania. This paper presents an evaluation of 20 accessions, belonging to 7 species, collected from polluted or degraded lands in Oltenia. The intra- and interspecific variability, analyzed through the prism of the production characters for SRC, is significant. The study gives the possibility of selecting some valuable genitors, for a breeding strategy and creation of new forms more adapted to the new environmental challenges. The best results regarding biomass production, sprouting capacity and growth rate were recorded by some accessions, belonging to the species S. triandra, S. viminalis, S. fragilis.

Key words:*biomass, Salix sp. accessions, short rotation coppice, variability.*

INTRODUCTION.

The genus *Salix* L. (*Salicaceae*) includes more than 500 species and their natural hybrids, spread in the Northern Hemisphere. Many of these species are suitable for biomass production or environmental protection.Willow has been used since ancient times for its curative properties, due to its salicylic acid content, Hippocrates (400 BC) mentioning its analgesic and antifever properties. The use of willow for braids, due to the elasticity of the shoots, is mentioned by Herodotus (500 BC) (Karp et al, 2014). For some peoples (Celtic and Middle Eastern culture) it had a sacred meaning, being associated with death and rebirth (Karp et al., 2011). These uses are still valid today, but the consideration for biomass production started after 1970, with the oil crisis.

The SRC willow is popular in the countries of Northern Europe, North America, Canada, India, New Zealand, Japan (Kuzovkina et al., 2008), and more recently in the countries of Central and Eastern Europe (Poland, Hungary, Romania (Fisher et al., 2005;Panita et al., 2017), China (Wang et al., 2017). The willow's capacity for phytoaccumulation and/or phytodegradation of soil and water contaminants, due to its increased tolerance to heavy metals, inorganic salts and xenobiotics, makes it possible to establish crops on degraded lands (waste dumps, ash, marginal lands) (Drzewieckaet al., 2012; Iori et al., 2015;

Corneanu et al, 2018; Celma et al., 2022). In the last fifteen years, the interest for willow short rotation coppice in Romania increased.The clones certified in Romania are of Swedish origin, with high productivity, but this is not fully expressed, due to the climatic conditions and the attack of diseases and pests (Buzatu-Goanta et al., 2021), conditions that are not faced in the country of origin.

The study of the local *Salix* sp. germplasm is of interest for a future breeding program and obtaining hybrids with resistance to drought, diseases and pests.Native species of willow have been taken into account as a source of resistance genes in breeding programs by many researchers(Kopp et al., 2001; Iori et al., 2015, Stolarski et al., 2020).

The aim of this study was to evaluate the potential of 20 willow accessions, in order to be used in land reclamation or to be used as genitors in a breeding program in different sites, even marginal ones.

MATERIALS AND METHODS

Biological material. The20 willows accessions were collected from the Oltenia area, Romania, in February 2015 (Table 1).

Table 1. The collectionlocation of the willow accessions

Field No.	Specie	Location
20	*Salix alba*	Station ICPA
21	*Salix fragilis*	Pocruia1
22	*Salix alba*	Diculesti
23	*Salix alba*	Sohodol
24	*Salix fragilis*	Pocruia 2
25	*Salix alba*	Bobicesti 232
26	*Salix fragilis*	Bobicesti P1
27	*Salix fragilis*	Bobicesti P2
28	*Salix purpurea*	Bobicesti P3
29	*Salix babylonica*	BobicestiA
30	*Salix pentandra*	CSD Peșteana 1
31	*Salix triandra*	CSD Peșteana 2
32	*Salix fragilis*	CSD Fărcășești 1
33	*Salix alba*	CSD Fărcășești 2
34	*Salix pentandra*	CSD Fărcășești 3
35	*Salix alba*	Negomir
36	*Salix fragilis*	CSD Pinoasa 1
37	*Salix fragilis*	CSD Pinoasa 2
38	*Salix purpurea*	Pocruia2
39	*Salix viminalis*	BobicestiP4
CSD : coalsterile dump		

The collection of accesssions was established in March 2015 in Experimental Didactic Station of the University of Life Sciences ”King Michael I” from Timisoara(Western Plain, elevation - 88m, N 45047'06”, E 21012'50”, pH = 6.1, cambic chernozem soil, precipitation - 592 mm) (Hernea et al., 2016).

Working method : The willow cuttings, 20 cm lenght have been planted in twin rows 70 cm apart and with 140 cm between each set of twin rows. 46 cuttings from each genotype were established in prepared soil at a distance of 80 cm between them(Hernea et al., 2016). Weed control was performed by herbicide and mechanical and manual works. No fertilizer or pest/disease pesticides were applied. After the first growing season, one row from each genotype was cut back.At the end of the first growing season, the survival rates were calculated.

There were performed biometrical observations regarding: number of shoots per stool; the maximum height of the main shoot, using a measuring pole (precision cm); the diameter at the base of the shoots usingan electronic caliper (0.01mm precision). In order to determine the biomass, in February 2016, all specimens from one of the two rows of each genotype in the collection were harvested. The mass of the specimens was determined by weighing with analytical balance. For this purpose every second row of each clone/hybrid was harvested and moisture was determined using the moisture -meter Voltcraft-300.Significance.

Data processing was performed withSoft STATISTICA 10.0 for Windows, Basic statistics (mean and standard deviation (SD), ANOVA/MANOVA, Duncan multiple range test for significance of the differences.

RESULTS AND DISCUSSIONS

The local genotypes of *Salix* sp., harvested from the spontaneous flora (from degraded lands, with a high content of heavy metals) possess heavy metal resistance genes, which give them natural tolerance to different stress factors. The importance of stress resistance genes of native genotypes was also highlighted by Huang et al. (2020) in a comparative salinity tolerance experiment using 36 genotypes from Canada, commercial and native hybrids. In order to obtain some valuable hybrids, which include the germplasm of productive North European hybrids and local germplasm with resistance genes, it was necessary to establish a collection of potential genitors.The 20 genotypes were collected after three collection expeditions in Gorj, Valcea and Olt counties, Oltenia region.

The 20 studied genotypes belong to 8 species. Biomass, height and yield provide a description of overall plant performance in different conditions, experimental or natural (Huang et al., 2020). That is why these characters were observed and analysed. The analysis of the variant reveals the fact that both the species and the genotype, in particular, have a very significant effect on the characters involved in production (Table 2). The values of the Fisher coefficients demonstrate the genetic determinism of these characters and once more the need to have a source of strong genes.The management practice obviously has a decisive role in amplifying the sprouting capacity of the stool.

The analysis of the significance of the difference allowed the classification of the analysed genotypes into size groups, by characters (Table 3). The highest amount of dry biomass was recorded for genotype 31.*Salix triandra*) (58.62 g/plant), with a very significant difference compared to all other genotypes. A second group of genotypes (7) with good production (b) had biomass between 24.32 g/plant (39. *S. viminalis*) and 32.47 g/plant (30. *S. pentandra*).

Table 2. **The effect of specie, genotype and management practice on the main traits – analysis of variance (Fisher test)**

Analysis of Variance (Marked effects are **significant at p < 0.05000**)				
Factor	Trait	F	p	Significance
Specie	Fresh biomass (g/plant)	**6.44226**	**0.008932**	**
	Dry biomass (g/plant)	**5.73526**	**0.016926**	*
	Maxim height(cm)	**15.11312**	**0.000000**	***
	Shoots no./stool	**10.99544**	**0.000000**	***
	Basis diameter (mm)	**12.80553**	**0.000000**	***
Genotype	Fresh biomass (g/plant)	**20.46641**	**0.000000**	***
	Dry biomass (g/plant)	**18.62310**	**0.000000**	***
	Maxim height (cm)	**27.68809**	**0.000000**	***
	Shoots no./stool	**9.61861**	**0.000000**	***
	Basis diameter (mm)	**10.07029**	**0.000000**	***
Management practice (uncut/cutback)	Fresh biomass (g/plant)	**7.07903**	**0.000000**	***
	Dry biomass (g/plant)	**4.65934**	**0.000000**	***
	Maxim height (cm)	**47.3181**	**0.000000**	***
	Shoots no./stool	**156.0765**	**0.000000**	***
	Basis diameter (mm)	**193.6941**	**0.000000**	***
All factors	Fresh biomass (g/plant)	**21.72943**	**0.000000**	***
	Dry biomass (g/plant)	**20.37132**	**0.000000**	***
	Maxim height (cm)	**19.57314**	**0.000000**	***
	Shoots no./stool	**20.08220**	**0.000000**	***
	Basis diameter (mm)	**18.54661**	**0.000000**	***

Table3 Productivity of willow accessions after two growing seasons according with genotypes and management practice (uncut and cut back) after the second growth season and biomass production after one growing season

Genotype	Biomass (g/plant) mean			Maxim height (cm) mean				No shoots/ stool mean			
	Fresh	Dry (10%)	DT	Uncut	DT	Cut back	DT	Uncut	DT	Cut back	DT
20.Salix alba	105.74	25.00	bc	269.72	ba	252.38	b	3.50	b	7.65	bc
21.Salix fragilis	82.14	17.51	cb	207.39	c	193.68	c	1.63	c	9.39	b
22.Salix alba	72.90	15.12	c	288.92	ba	291.67	a	2.17	c	3.22	c
23.Salix alba	64.49	13.00	c	254.09	bc	222.27	b c	2.95	bc	7.73	c
24.Salix fragilis	142.00	27.05	b	240.00	bc	202.41	c	3.91	b	11.86	a b
25.Salix alba	56.75	12.34	c	208.16	c	164.64	d	3.26	b	5.36	c
26.Salix fragilis	128.27	25.25	bc	234.57	bc	224.09	b c	3.65	b	14.95	a
27.Salix fragilis	127.71	27.00	b	255.56	bc	249.50	b	2.78	bc	9.50	b
28.Salix purpurea	56.43	12.54	c	192.50	c	183.48	c	2.09	c	9.52	b
29.Salix babylonica	110.51	24.67	bc	319.50	ab	223.61	b c	1.85	c	5.44	c
30.Salix pentandra	163.22	32.47	b	295.29	ba	223.41	b c	3.00	b	6.23	b c
31.Salix triandra	277.90	58.62	a	344.30	a	298.57	a	2.13	c	5.67	c
32.Salix fragilis	45.24	11.37	c	191.19	cd	157.86	d	2.00	c	5.00	c
33.Salix alba	30.27	6.23	d	146.92	d	145.63	d	2.62	b	3.38	c
34.Salix pentandra	30.07	6.19	d	153.00	d	145.00	d	3.40	b	4.75	c
35.Salix alba	37.10	7.88	d	162.27	d	141.36	d	2.45	bc	3.73	c
36.Salix fragilis	46.62	9.23	d	162.06	d	158.18	d	1.88	c	7.55	b
37.Salix fragilis	27.58	6.05	d	211.47	c	130.31	d	2.18	c	2.00	c
38.Salix purpurea	52.91	11.89	c	147.50	d	138.00	d	3.38	b	4.60	c
39.Salix viminalis	110.92	24.32	bc	242.22	bc	217.67	b c	6.89	a	12.27	a b
All Grps	82.40	17.21		211.41		189.32		3.57		8.79	

DT = Different letters between genotypes denote significant differences (Duncan test, $p < 0.05$).
(a = the highest value; b = significant < a; c significant < b; d significant < c)

Table 4 Interspecific variability -the mean values per specie, depending on the management practice, of the analyzedtraits

Specie	UC	No of shoots/stool				Basis diameter (mm)				Shoot height (cm)			
	C	Mean	SD.	DT1	DT2	Mean	SD	DT1	DT2	Mean	SD	DT1	DT2
Salix alba	UC	2.88	0.14	b	b	28.74	1.24	bc	a	212.38	7.13	b	a
Salix fragilis		2.62	0.16	b	b	33.38	1.09	b	a	213.16	4.81	b	a
Salix purpurea		2.63	0.26	b	b	27.79	1.50	cb	a	173.55	5.89	c	a
Salix babylonica		1.85	0.41	b	b	33.30	2.04	b	a	319.50	8.00	a	a
Salix pentandra		3.33	0.37	b	b	18.81	0.73	d	a	243.15	16.37	b	a
Salix triandra		2.13	0.30	b	b	49.94	2.08	a	a	344.30	17.65	a	a
Salix viminalis		6.89	0.53	a	b	20.36	1.13	d	a	242.22	6.67	b	a
Salix alba	C	5.21	0.31	c	a	21.46	0.73	a	b	197.69	6.57	bc	a
Salix fragilis		9.03	0.56	b	a	20.26	0.57	a	b	187.73	4.49	cb	a
Salix purpurea		7.58	0.59	b	a	19.62	0.90	a	b	165.53	5.92	c	a
Salix babylonica		5.44	0.63	c	a	18.11	1.32	a	b	223.61	8.69	b	b
Salix pentandra		6.23	1.19	c	a	15.17	0.80	ba	a	223.41	8.79	b	a
Salix triandra		5.67	0.43	c	a	19.88	1.87	a	b	298.57	11.94	a	b
Salix viminalis		12.27	1.31	a	a	16.43	1.04	ab	a	217.67	10.14	b	a

DT1 = Duncan test – the significance of the differences between specie,
DT2 =Duncan test – the significance of the differences between management practice (UC = uncut; C = cut - back) the general mean per specie (a = the highest value; b = significant < a; c significant < b; d significant < c)

Biomass evaluation after the first growing season is not relevant for producers, but it can provide valuable information for the selection of genitors for hybridization. In the second growing season, plant growth was luxuriant, due to the special climatic conditions for Banat in July-August 2016, especially rainy months. The soil has low permeability, so that throughout the two months, the water has stagnated, in a constant layer of 30-40 cm.

The height of the plants recorded high values for 8accessions, which had growths of over 200 cm, most exceeding the average of the experiment. Harvesting the shoots in February 2016 stimulated a strong shooting and vigorous growth (Table 3). The maximum number of shoots was recorded for 26. *S. fragilis*(14.95 shoots/stool) andand39. *S. viminalis*(12,27 shoots/stool), which do not have spectacular growth, but are suitable for culture on soils that must be fixed, against shattering (such as, for example, ash deposits).The significant differences between the genotypes of the same species appear due to the high individual variability they present, a fact also observed by Andleeb et al. (2011), when studying the molecular polymorphism of 4 genotypes of *S. viminalis* from India.

Interspecific variability. The interspecific variability was evaluated by comparing the general average values per species, taking into account the cultural method. The significance of the difference was achieved with the Duncan multiple range test, for $p<0.05$ (DT1). Also, the general means were compared, for each species, for the management practice applied (DT2; Table 4). The sprouting capacity, an important parameter of biomass production, increased significantly after cut-back, the average values of all species being significantly higher in the cut-back variant, compared to uncut. The best results were recorded with *S. viminalis* (12.27± 1.31 shoots/stool in the cut-back variant and 6.89± 0.53 shoots/stool in the uncut variant. In the uncut experimental variant there are no significant differences between species, excluding *S. viminalis*, with the highest value (a), the number of shoots / stool being between 1.85±0.41 (*S. babylonica*) and 3.33± 0.37 (*S. pentandra*).After applying the cut-back, two value groups are highlighted, group b, which includes 7.58-9.03 shoots/stool (*S. purpurea*, respectively *S. fragilis*). Group C brings together the remaining 4 species, with values between 5.21 (*S. alba*) and 6.23 shoots/stool (*S. pentandra*).

The diameter at the base of the main shoot recorded values between 18.81±0.73 mm (*S. pentandra*) and 49.94±2.08 mm (*S. triandra*), the species being divided into 4 value groups. It is noteworthy that in the cut – back variant, the differences between the species, regarding the basal diameter, are insignificant. In the species *S.pentandra* and *S.viminalis*, there are no significant differences between the uncut - cut-back variants.

For height growth, the highest values were recorded for *S.triandra*, which proved to be the most robust species, with a fast growth rate, in both experimental variants (344.30 cm -uncut and 298.57 cm -cut - back). The lowest growth values were noted for *S. purpurea*, in both experimental variants. In general, with the exception of *S. triandra* and *S. babylonica*, there are no significant differences between the two experimental practices.

The differences between the species are significant, for the sprouting process, after cut-back, and regarding the growth rate, the differences are significant in the uncut variant and faded in the cut-back variant. The highest sprouting capacity was observed in *S. viminalis*, a species that is otherwise found in many commercial hybrid combinations. The most robust species turned out to be *S. triandra*, with a more modest sprouting capacity, but for biomass production it takes over the rapid growth rate, due to its rich foliage and a high photosynthetic rate. Andralojc et al. (2014), in a study on the correlations between various parameters of photosynthesis and growth found that in *S. triandra*, which supports higher concentrations of CO_2 in the environment, assimilation is at a significant advantage. The analysis of molecular polymorphism revealed that associated Rubisco large subunit (LSU) genes in *S. triandra* is different from other genotypes, contained three differrent codons, representing amino-acidsubstitution (Andralojc et al., 2014). *S. triandra* is mentioned, as a species of particular interest, distinct from most willow species (Trybush et al., 2008).

Establishing sex and fertility of the accessions. The genus *Salix* is a polyploid complex, which raises many taxonomic problems due to easy interspecific hybridization. The basic number of chromosomes of the genus x = 19.

The collection of genitors from Oltenia, includes 20 genotypes, belonging to 7 species, and the research during two seasons of vegetation was carried out so that a pre-selection of them in terms of resistance to stress conditions (unirrigated, unfertilized culture, without treatments to combat diseases and pests) was performed. Of the 20 genotypes, 9 are female, 11 are male, because willows are dioecious (Table 5). Salix has a ZW sex-determining system in whichfemales are primarily the heterogametic sex (ZW), as Gouker mention in its studies. Gouker et al. (2021) noted in the studies carried out on S. purpurea, the fact that there is a sexual dimorphism, in this species, regarding the production capacity of the male, apparently due to the ability to accumulate more nitrogen. Cytology studies have shown that all male genotypes have viable, fertile pollen. Female clones were pollinated (free pollination) and formed viable seed. Hybrid plants obtained from seeds have been grown in pots and are viable.

All 9 genotypes with female plants are able to be hybridized with male parents, even with different degrees of ploidy, in this case resulting in plants with odd degree of ploidy, sterile. The multiplication of plants is done vegetatively (through cuttings, sterility is not a problem for the establishment of crops. Female genitors particularly valuable for biomass production are clones 30 and 31.

Table 5.The sex and ploidy level (PL) of the potential genitors

Female genotypes	2n	PL	Male genotypes	2n	PL
22.Salix alba	*76*	*4x*	*20.Salix alba*	*76*	*4x*
21.Salix fragilis	*76*	*4x*	*26.Salix fragilis*	*76*	*4x*
23.Salix alba	*76*	*4x*	*27.Salix fragilis*	*76*	*4x*
24.Salix fragilis	*76*	*4x*	*28.Salix purpurea*	*38*	*2x*
25.Salix alba	*76*	*4x*	*33.Salix alba*	*76*	*4x*
29.Salix babylonica	*38*	*2x*	*34.Salix pentandra*	*76*	*4x*
30.Salix pentandra	*76*	*4x*	*35.Salix alba*	*76*	*4x*
31.Salix triandra	*38*	*2x*	*36.Salix fragilis*	*76*	*4x*
32.Salix fragilis	*76*	*4x*	*37.Salix fragilis*	*76*	*4x*
			38.Salix purpurea	*38*	*2x*
			39.Salix viminalis	*38*	*2x*

CONCLUSIONS

Biometric observations revealed a large intra- and interspecific variability of the genotypes of *Salix* sp. collected from Oltenia and cultivated in the Western Plain of Banat.

The analysis of the variance revealed that both the effect of the genotype is very significant on all production characters, as well as the effect of the species. The management practice (cut-back/uncutted) has a very significant effect on shooting capacity

After testing in comparative crops, some of the genotypes could be certified: clone 26 *S. fragilis*, 30 *S. pentrandra* and clone 31 *S. triandra* for short-rotation coppice, both under normal soil-climatic conditions and on degraded soils.

Valuable clones, for different traits, will be introduced into a breeding program, with a certain strategy, if financial resources will allow this study.

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REFERENCES

Andleeb, L., Munshi, A. H., & Dar, A. R. (2011). Genetic Diversity in *Salix viminalis* in the Kashmir Valley, India. American journal of Biochemistry and Molecular Biology, 2011: 1-7.

Andralojc, P. J., Bencze, S., Madgwick, P. J., Philippe, H., Powers, S. J., Shield, I., ... & Parry, M. A. (2014). Photosynthesis and growth in diverse willow genotypes. Food and Energy Security, 3(2), 69-85.

Buzatu-Goanta, C., Corneanu, M., &Babeanu, C. (2021). Salix Accessions with Potential For New Hybrids. A Case Study from Banat Area. Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering, 10, 214-220.

Celma, S.; Sanz, M.; Ciria, P.; Maliarenko, O.; Prysiazhniuk, O.; Daugaviete, M.; Lazdina, D.; von Cossel, M.(2022) Yield Performance of Woody Crops on Marginal Agricultural Land in Latvia, Spain and Ukraine. *Agronomy* **2022**, *12*, 908. https://doi.org/10.3390/agronomy12040908

Corneanu, M., Corneanu, C. G., Craciun, C., Netoiu, C., Hernea, C., &Cojocaru, L. (2018). The potential of two willows genotypes for radionuclide and heavy metals accumulation. A case study of Rovinari coal ash pond (GorjCounty, Romania). Annals of the University of Craiova-Agriculture, Montanology, Cadastre Series, 48(2), 58-70.

Drzewiecka, K., Mleczek, M., Gąsecka, M., Magdziak, Z., &Goliński, P. (2012). Changes in *Salix viminalis* L. cv.'Cannabina'morphology and physiology in response to nickel ions–Hydroponic investigations. Journal of Hazardous Materials, 217, 429-438.

Fischer, G., Prieler, S., & van Velthuizen, H. (2005). Biomass potentials of miscanthus, willow and poplar: results and policy implications for Eastern Europe, Northern and Central Asia. Biomass and Bioenergy, 28(2), 119-132. https://doi.org/10.1016/j.biombioe.2004.08.013

Hernea, C., Corneanu, M., Sarac, I., &Turcu, E. (2016).The potential of Romanian and Swedish willow forshort rotation coppice in the specific condition of Banat plain: a comparative analyze. South WesternJournal of Horticulture, Biology and Environment,7(2), 115-126.

Xinyi Huang, Raju Y Soolanayakanahally, Robert D Guy, Arun S K Shunmugam, Shawn D Mansfield, Differences in growth and physiological and metabolic responses among Canadian native and hybrid willows (Salix spp.) under salinity stress, Tree Physiology, Volume 40, Issue 5, May 2020, Pages 652–666, https://doi.org/10.1093/treephys/tpaa017

Gouker, F. E., Carlson, C. H., Zou, J., Evans, L., Crowell, C. R., Smart, C. D., ... & Smart, L. B. (2021). Sexual dimorphism in the dioecious willow *Salix purpurea*. American Journal of Botany, 108(8), 1374-1387.

Iori, V., Pietrini, F., Massacci, A. and Zacchini, M., (2015) Morphophysiological responses, heavy metal accumulation and phytoremoval ability in four willow clones exposed to cadmium under hydroponics. In Phytoremediation, 87-98. Springer, Cham.

Karp, A., Hanley, S. J., Trybush, S. O., Macalpine, W., Pei, M., & Shield, I. (2011). Genetic improvement of willow for bioenergy and biofuels free access. Journal of integrative plant biology, 53(2), 151-165.

Karp, A. (2014). Willows as a source of renewable fuels and diverse products. Challenges and Opportunities for the World's Forests in the 21st Century, 617-641.

Kuzovkina, Y.A., M. Weih, M.A. Romero, J. Charles, S. Hurst, I.McIvor, A. Karp, S. Trybush, M. Labrecque, T.I. Teodorescu, N.B.Singh, L.B. Smart, and T.A. Volk. 2008. Salix: Botany and GlobalHorticulture. Horticultural Reviews 34:447–489.

Nord-Larsen, T.; Sevel, L.; Raulund-Rasmussen, K. (2015) Commercially grown short rotation coppice willow in Denmark: Biomass production and factors affectingproduction. BioEnergyRes. 2015, 8, 325–339

Panita, O., Soare, M., Corneanu, M., &Iancu, P. (2017). The variability study of morphological characters on some *Salix* spp. genotypes. Annals of the University of

Craiova-Agriculture, Montanology, Cadastre Series, 47(2), 209-216.Scarlat, N., Dallemand, J.,Taylor, N., Banja, M., Sanchez L.J. , Avraamides, M. (2019). Brief on biomass

Stolarski, M.J.; Krzyżaniak, M.; Załuski, D.; Tworkowski, J.; Szczukowski, S. (2020) Effects of Site, Genotype and Subsequent Harvest Rotation on Willow Productivity. *Agriculture* 2020, *10*, 412. https://doi.org/10.3390/agriculture10090412

Trybush , S. , S. Jahodova , W. Macalpine and A. Karp . 2008 .Genetic studies of a germplasm resource reveal newinsights into relationships among *Salix* subgenera, sectionsand species .BioEnergy Res. 1 : 67 – 79 .