RESEARCHES CONCERNIG THE GROWING PHISIOLOGY OF SOME SALIX SP. GENOTYPES

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ABSTRACT

Willow is a species that can be grown for biomass in areas with colder temperatures. This culture gives good results in regions with optimal amounts of water level. Recent studies are focused on expanding willow in southern warmer areas, but the culture can be affected by water stress and heat.

In this regard, research has tracked the behavior of a range of native and foreign *Salix* sp. genotypes in the climatic conditions of Southern Romania. In this area the culture is characterized by high temperature variations and long periods of lack of rainfall.

It was aimed the analysis of the physiological processes (intensity of photosynthesis, transpiration intensity) of physiological indices (stomatal conductance, total chlorophyll, total water and dry substance) as well parameters with a direct influence on them (photosynthetic active radiation, leaf temperature). Also, it was recorded growth capacity and the number of shoots.

Analysis of physiological indices allowed the assessment of the behavior of genotypes under stress and thermal fluid and highlighted those with good adaptability to the climatic conditions of the studied area which makes a larger amount of biomass.

Keywords: willow, drought stress, photosynthesis, adaptability

INTRODUCTION

Salix spp. is a clean source of energy and a way for biorecovering degraded fields, becoming very popular and giving benefits to social and environmental areas. Also it is used as raw material for chemical process and manufacturing industry [1]. Using renewable energy sources reduces the dependence on fossil fuels and reduces emissions of greenhouse gases [2]. The willow crop in SRC is considered a sustainable source of biomass with a positive greenhouse gas (GHG) due to their potential to fix carbon (C) in soil [3].

Salix spp., is a fast growing species and the crop is suitable for the biomass yield. The economic efficiency of this species is given by the high level of biomass yield. It can also be used as a proper crop for soil phytoremediation due to its potential of absorbing heavy metals.

On the other hand, willow crops offer some disadvantages such as long period for recovering the initial investment because of its long period of time until it produces the biomass, variability of climatic factors and the variety of high technologies [1].

Bioenergy production based on short rotation coppice plantations (SRC) is widely introduced in some European countries, USA and Canada [4]. Willows have a special place among energy crops due to their high potential in productivity and broad tolerance to environmental factors [5].

The researches conducted by various authors at *Salix* have focused on the influence of climatic factors on the physiology of different genotypes, the necessary minerals and biomass.

The key factor for willow production is the water regime of plants. Willows belong to the group of phreatophytes, having an increased demand for water during vegetation. Therefore, the focus on water regime at willow plants is highly recommanded in the investigation. Water retention ability represents a physiology indicator related to water use efficiency [5]. It was found that diurnal dynamics of transpiration showed one peak (typically at midday) for all clones [5] and by measuring the net photosynthesis of *Salix* during the period between May and October in central Scotland, the greatest rates were found in July and September [6].

Within all plants there exists a functional equilibrium of biomass, where additional biomass is allocated to an organ to take up the resource that is most limiting growth [7]; [8]; [9]. An understanding of these principles can be found in plant ecology, but they have many applications in agricultural research as allocation sets limits on biomass production and utilisation [8]. Allocation patterns can be affected by numerous environmental factors to varying extents including: light, nutrients, water, elevated atmospheric CO_2, temperature, salinity and mechanical perturbation, and have been reviewed by numerous authors using a plethora of data from environmental manipulation studies [8]; [9]; [10]; [11].

In Romania, there have been studied and evaluated clones of Salix spp. regarding yield potential of biomass for clean energy. Between 2009-2013, the research of the energy and yield potential of local and foreign varieties of willow was made by SCDP Govora Valcea, in the northern region of Oltenia, Romania, which identified four native genotypes with high yields of biomass in comparison with foreign genotypes [12].

In this study, the research followed the behavior of a range of genotypes of *Salix spp.*, in climatic conditions from southern Romania, the area experiencing high temperature variations and periods of drought.

MATERIALS AND METHODS

The study was conducted on an assortment of genotypes of Salix spp., in the climatic conditions from southern Romania, on the fields between Radovan and Fântânele village, Dolj County, with the GPS coordinates: N 44°10'05", E 23°36'13", Altitude 93 m. The soil have moderate content of humus 2.31%, with pH value 6.24.

The biological material was represented by several Romanian and foreign genotypes and after examining the behavior of those, there were revealed nine genotypes that can be suitable for cropping in the conditions in this area, which are: RO892, RO1077, RO1082, Tora, Fragisal, Pesred, Robisal, Inger, Olof.

The experiment was designed according to randomized complete block split-block with three replications, applying conventional crop technology of energy willow.

The genotype was the only analyzed factor, data collected were statistically analyzed with ANOVA using the Fisher's technique. The least significant difference (LSD) test at 0.05 was used to compare means.

Regarding the climatic conditions during the growing season (2016), there were recorded temperature oscillations, very dry periods or excess rainfall in a short time (Fig 1).

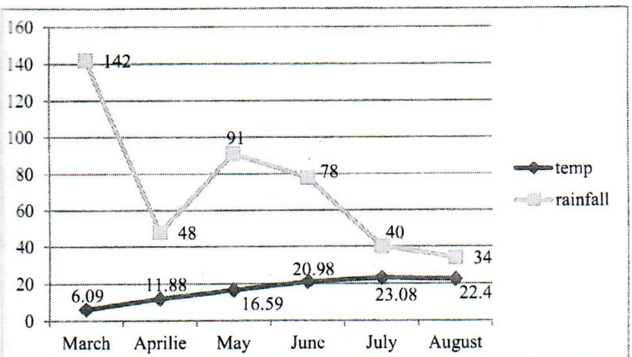


Fig. 1. Variation of monthly average temperatures (^{0}C) and rainfall (mm) during March -August/2016 in the area of experimentation

In July and August rainfall was low and the temperatures were very high, which strongly influenced the growth of plants.

Physiology research consisted of analyzing the physiological processes (intensity of photosynthesis, intensity of transpiration), some physiological indices (stomatal conductance, total chlorophyll, dry mater), and the parameters directly acting on them (photosynthetically radiation, active the temperature of the leaf). Also it was recorded the growth capacity, the number of shoots.

Photosynthesis and transpiration intensity, photosynthetic active radiation (PAR), leaf temperature, stomatal conductance were measured using portable analyzer Lci (ADC BioScientific Ltd, UK) with the microchamber assay, the surface of which amounts to 6.25 cm^2. Chlorophyll content was determined using the Minolta SPAD 502 portable chlorofilometer.

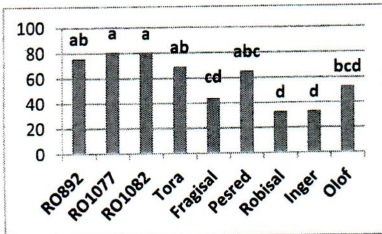
The total content of water and dry matter was determined by gravimetric methods, using the oven (drying of plant material at a temperature of 100-105^{0}C).

RESULTS AND DISCUSIONS

Analyzing obtained data on the morphology of growth, shown in Fig. 1 and Fig. 2, it can observed high variability of the studied genotypes. Concerning the height of the plants, it was recorded amplitude variation of 47.63 cm, with a maximum on RO10 genotype and a minimum on Inger genotype.

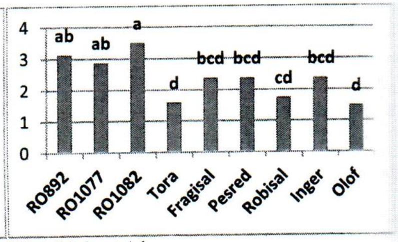
The first two genotypes classified respectively in RO1077 and RO1082 quantif significant differences from the last four genotypes classified respectively Olof, Frag Inger and Robisal. Incidentally, the last two genotypes classified records signific negative differences from the first five genotypes classified.

Average length of annual growth was strongly influenced by drought installed in J and August. Under northern Oltenia (Romania) conditions, where the rainfall regim much richer and cooler temperatures during the vegetation period, annual increa recorded higher values to some of the genotypes studied in this experiment.



LSD 5%= 24.41 cm

Fig. 2 The average lenght of annual growth (cm);



LSD 5%=1.04 shoots/pl

Fig. 3 No. of shoots at studied *Salix* genotyp

Concerning the number of shoots, it was recorded a variation amplitude of shoots/pl., with a maximum on RO1082 (3.7 shoots/pl) genotype and a minimu Olof genotype. First classified genotype obtains significant differences compare the last six classified genotypes, while the last classified genotype records ne significant differences compared with the first three genotypes classified Fig. 3.

The evaluation of biomass production potential of eight willow varieties (*Sal* grown on arable land in the soil and climatic conditions of south-western Slovak number of shoots per plant, in the second year, after the cutback ranged from 5 13.28 shoots [2].

The results obtained on some physiological indices are presented in Table 1. Fr analysis of data obtained in pedo-climatic area of research, on the analyzed clon highest value of the intensity of photosynthesis (11.54 $\mu mol/m^2/s$) was recor genotype Olof, wich registered a photosynthetically active radiation value of $\mu mol/m^2/s$, on temperature of 31.6^{0}C of the leaf and a stomatal conductance va 0.07 $mmol/m^2/s$.

High levels of photosynthesis intensity were recorded on Pesred genotype $\mu mol/m^2/s$) and Robisal (9.34 $\mu mol/m^2/s$), on the same conditions of temperat light intensity.

The lowest values were recorded at RO892 genotype (2.41μmol/m^2/s), on a photosynthetic active radiation 1154 μmol/m^2/s, temperature of 28.2^{0}C and leaf stomatal conductance of 0.09 mmol/m^2/s. Low values were also recorded on RO 1082 genotype (2.69 μmol/m^2/s) and Fragisal (3.19 μmol/m^2/s).

On the other analyzed variants were recorded average values of the intensity of photosynthesis between these limits.

Table 1. The variation of physiological characters at *Salix* genotypes studied

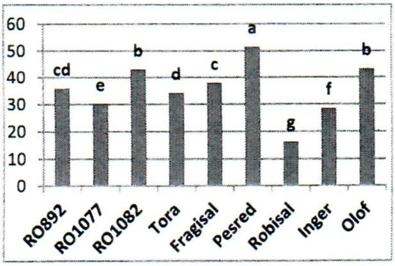
Caract. / Clone	Photosynthesis (μmol/m^2/s)	Intensity transpiration (mmol/m^2/s)	Photosynthetically radiation (μmol/m^2/s)	Active temperature of the leaf (^{0}C)	Stomatal conductance (mmol/m^2/s)
RO892	2.41+0.37i	1.12+0.12f	1154+102.68d	28.20+0.71c	0.09+0.01cd
RO1077	4.54+0.58h	1.37+0.19e	1080+74.09e	28.00+0.93c	0.08+0.01de
RO1082	2.69+0.47f	1.61+0.22cd	1038+28.13e	27.80+0.73c	0.11+0.02b
Tora	3.89+0.49g	1.58+0.23d	1226+87.41bc	28.00+0.75c	0.09+0.01cd
Fragisal	3.19+0.39i	1.15+0.20f	1185+58.06cd	28.10+0.58c	0.08+0.01de
Pesred	10.58+0.60d	1.71+0.17bc	1335+65.30a	28.50+0.57c	0.10+0.02bc
Robisal	9.34+0.22c	1.79+0.14b	1189+67.38cd	28.40+0.46c	0.13+0.01a
Inger	6.75+0.35b	2.14+0.19a	1232+72.30bc	36.00+1.85a	0.11+0.01b
Olof	11.54+0.26a	1.36+0.16e	1275+70.71ab	31.60+1.08b	0.07+0.01e
LSD 5 %	0.4	0.18	69.82	1.65	0.02

Regarding the leaf transpiration, the highest intensity (2.14 mmol/m^2/s) it was recorded on Inger variants, with a photosynthetic active radiation value of 1232 μmol/m^2/s, a leaf temperature of 30.6^{0}C and a value of stomatal conductance of 0.11 mmol/m^2/s.

High levels of leaf transpiration were recorded on Pesred genotype (1.71mmol/m^2/s) and Robisal genotype (1.79 mmol/m^2/s). The lowest value of the intensity of transpiration was recorded on RO 892 genotype (1.12 mmol/m^2/s), with a photosynthetic active radiation value of 1154 μmol/m^2/s, a leaf temperature of 30.6^{0}C and a value of stomatal conductance of 0.09 mol/m^2/s.

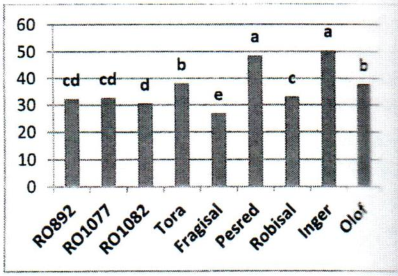
Low values were also recorded on Fragisal genotype, (1.15 mmol/m^2/s), on a value of stomatal conductance of 0.08 mmol/m^2/s. On the other variants were recorded average values of the intensity of transpiration between 1.36-1.50 mmol/m^2/s. According to recorded data, RO892 genotype has the lowest values of photosynthesis and transpiration.

Regarding the content of chlorophyll, it showed higher values in Pesred genotype, 51.5 SPAD units, high value correlates with the intensity of photosynthesis (Fig.4).



LSD 5%=2.7 SPAD units

Fig. 4. The content of chlorophyll;



LSD 5%=2.28 g/100 g

Fig. 5. The content of soluble dry substance

The lowest chlorophyll content was recorded on Robisal genotype (16.23 SPAD units) and on the other clones the values were between 22.6 and 43.0 SPAD units.

In the climatic conditions in the south of Romania, the dry matter content of the leaves showed the highest values on Inger genotype (50.43%) and Pesred (48.53%) (Fig. 5).

High values for total water content on Fragisal genotype may be due to capacity to reduce water losses by reducing the openness of the stomata, evidenced by low levels of transpiration intensity.

Photosynthesis intensity values are correlated with photosynthetic active radiation values, stomatal conductance and amount of chlorophyll.

Higher levels of intensity of transpiration may be an adaptive advantage for a good supply of water as transpiration generates a suction force that ensures the rise of a greater amount of water in the plant body. In atmospheric and soil drought conditions appropriate to experimentation area, high intensity values of transpiration, if not correlate with an increased photosynthesis, lead to dehydration and reduced plant productivity. From this point of view, are considered better suited the clones that have increased the intensity of photosynthesis and low transpiration intensity. On the clones that develop high values of transpiration intensity, but low levels of photosynthesis intensity, we can say that water is used inefficiently.

CONCLUSION

Global energy demand is becoming greater and renewable energy is an important direction for researchers. The production of bioenergy will become a priority, and energy crops for biomass, such as willow (like energy wilow) is a perspective to be followed in the future.

Test results of *Salix* spp. genotypes showed their behavior in the specific conditions in southern Romania and highlighted the differences in adaptation. Recorded values of the stem length and number of shoots/pl. were strongly influenced by the lack of rainfall and high temperatures in July and August and because of that, they were much diminished.

Concerning the physiological index, the highest value of the intensity of photosynthesis (11.54 µmol/m^2/s) was recorded on Olof genotype, followed by indigenous genotypes Robisal and Pesred. Regarding the leaf transpiration, the highest value (2.14 mmol/m^2/s) was recorded on Inger genotype, which causes the plants dehydration and reduces the production of biomass, and the lowest value was recorded on RO 892 genotype (1.12 mmol/m^2/s) followed by Fragisal (1.15 mmol/m^2/s). RO1077 and Olof genotypes have higher photosynthesis intensity and low transpiration intensity, and can be considered better adapted to local conditions.

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