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**WATER AND IONIC BALANCE
IN THE LEAVES OF BASKET WILLOW (*Salix viminalis* L.)
CULTIVATED IN HYDROPONICS
WITH DIFFERENT SALINITY LEVELS**

**BILANS WODNY ORAZ JONOWY
W LIŚCIACH WIERZBY WICIOWEJ (*Salix viminalis* L.)
UPRAWIANEJ W KULTURACH WODNYCH
O RÓŻNYM STOPNIU ZASOLENIA**

Abstract: In the study, a significant effect of different NaCl concentration in Hoagland's medium was showed on the water and ionic balance in the leaves of three clones of *Salix viminalis* L., ie 'Bjor', 'Jorr' and 'Tora'. The measurements of water indicators (RWC and WSD) as well as the content of monovalent (K⁺ and Na⁺) and bivalent (Ca²⁺ and Mg²⁺) cations in the leaves of basket willow clones allow to conclude that under high salinity of hydroponics with sodium chloride the 'Bjor' clone was characterised by more favourable water and ionic balance when compared with the 'Jorr' and 'Tora' clones, as well as by effective mechanism of decreasing the uptake of sodium ions in leaves under the highest salt concentration in the medium. The results indicate that the 'Bjor' clone is best adapted to survival under salt stress conditions.

Keywords: hydroponics, salinity, *Salix viminalis* L., water balance, ionic balance

The basket willow (*Salix viminalis* L.), know also as the energy willow, is characterised not only by a quick and large increase in biomass but also by a broad tolerance to unfavourable environmental conditions. Considering its specific genetic and physiological properties, this plant can be used both for energy production purposes and reclamation of anthropogenically degraded land [1–3]. At present, one of the main stressors for plants is substratum salinity induced by excessive fertilisation or NaCl use for glazed ice control. The most frequently used compound, and at the same time the most toxic one, is sodium chloride. The excess of sodium and chlorine ions in nutritive environment induces a decrease in water chemical potential in the soil solution,

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reducing the same its availability for plants. Furthermore, it disturbs ion economy and normal plant feeding, bringing about in addition the oxidative stress [4–6].

Due to little information in the available literature on the effect of salt stress on new forms of energy willow, studies were taken with the aim to determine the volume of changes in the water and ionic balance in three clones of *Salix viminalis* under the influence of different NaCl doses added to hydroponics.

Material and methods

The biological study material was three clones of basket willow (*Salix viminalis* L.): ‘Bjor’, ‘Jorr’ and ‘Tora’, coming from a plantation of the Department of Plant Physiology of the Agricultural University in Szczecin. In 2006–2007, a hydroponics experiment was carried out in the Vegetation Room of the Agricultural University in Szczecin under controlled conditions. The experiment was set up as a two-factor one in randomised complete block design in three series (ie in May, June and July) and in three replications. The first experimental factor was different NaCl concentration in Hoagland’s medium, ie 0.068, 0.136 and 0.170 mol NaCl · dm⁻³, and the control, which was a complete Hoagland’s medium, whereas three clones of *Salix viminalis* were the second one.

The willow cuttings were placed at first in appropriately prepared hydroponics, with a capacity of 2 dm⁻³ each (2 cuttings per each container), filled with complete Hoagland’s medium. Next, after about 14 days, when the cuttings rooted themselves and developed shoots, about 20 cm long, the mediums were replaced, differentiating in them the NaCl concentration, in conformity with to the experiment variants adopted.

After 72 hours from exposing the hydroponics to salinity, the water balance was determined in the leaves of examined willow clones basing on the *relative water content* (RWC) and *water saturation deficit* (WSD) indicators according to Bandurska [7]. Then, after 168 hours from exposing the medium to salinity, the content of selected macroelements, ie sodium, potassium, calcium and magnesium, was assayed in the collected and subsequently wet-mineralised leaves. The determination was made with atomic absorption spectrophotometry method (AAS).

Results referring to the content of mineral elements were processed statistically using two-factor analysis of variance. In order to determine the significance of differences between means for interactions, Tukey’s confidence semi-intervals were calculated at the significance level $\alpha = 0.05$ (LSD_{0.05}). In case of RWC, homogenous groups were determined, basing on the results given in grams. Considering the homogeneity of error variance, the synthesis of results from two years and three experiment series was performed.

Results and discussion

The salinity of medium with sodium chloride had a significant effect on the water balance of leaf tissues in two basket willow clones, out of three examined ones, which was measured by relative water content (RWC) and water saturation deficit (WSD) indicators (Table 1). The largest amount of water was a characteristic of the leaves of

'Tora' and 'Jorr' clones under control conditions (93.6 and 89.8 %, respectively) and in the lowest NaCl concentration (88.2 and 89.4 %, respectively). On the other hand, at the highest salinity of hydroponics, ie $0.170 \text{ mol NaCl} \cdot \text{dm}^{-3}$, the same clones showed a significantly higher water loss, up to 20 %, when compared with the 'Bjor' clone. In case of that particular clone, irrespective of NaCl concentration in the medium, the water balance in leaves was at a similar level, ie about 82–85 % RWC and 15–18 % WSD. This may be evidence of larger resistance of the 'Bjor' clone to relatively high salinity of medium with sodium chloride when compared with the 'Jorr' and 'Tora' clones, which responded under the same conditions with a rapid decrease in leaf tissue hydration.

Table 1

Relative water content (RWC) [%] and water saturation deficit (WSD) indicators in the leaf tissues of *Salix viminalis* L.

Clone	RWC [%]				WSD [%]			
	Concentration of NaCl in culture [$\text{mol} \cdot \text{dm}^{-3}$]				Concentration of NaCl in culture [$\text{mol} \cdot \text{dm}^{-3}$]			
	0.0 (Control)	0.068	0.136	0.170	0.0 (Control)	0.068	0.136	0.170
'Bjor'	82.4b	85.1b	84.0b	82.0b	17.6b	14.9b	16.0b	18.0b
'Jorr'	89.8a	89.4a	80.3b	61.7c	10.2a	10.9a	19.7b	38.3c
'Tora'	93.6a	88.2ab	84.9b	62.6c	6.4a	11.8ab	15.1b	37.4c

a, b, c – The values denoted by the same letter do not differ significantly at the level $\alpha = 0.05$.

The plant hydration is affected by many different factors, including the supply of mineral components to plants and how these compounds are managed. This is because the disturbance of ionic balance in medium, brought about by overabundance of Cl^- and Na^+ ions, induces a disturbance in the ionic balance in plants. According to Marosz [5], Starck et al [8] and Greszta et al [9] the salinity of natural environment with sodium chloride brings about changes in the cell metabolism since the absorbed sodium ions increase the hydrophilic ability of plasma colloids, which induces the binding of larger quantity of water. The potassium ions have a similar effect. This is explained by high hydration of leaf tissues observed in the examined willow clones at lower NaCl concentrations in the medium (Table 1).

In the carried out study, a significant increase in the content of Na^+ ions in the leaves of basket willow was found together with the increase of NaCl concentration in the medium, but only to $0.136 \text{ mol NaCl} \cdot \text{dm}^{-3}$ (Fig. 1). At the highest salinity, the content of sodium in the 'Jorr' and 'Tora' clones was maintained at the same level. On the other hand, the content of that element in the 'Bjor' clone decreased significantly. This attests to setting off a defensive mechanism by this willow clone, protecting it from absorption of excessive and harmful for cells quantity of Na^+ ions. The constant and invariable level of hydration in the 'Bjor' clone can be explained by this. On the other hand, a decrease in the hydration of the 'Tora' and 'Jorr' clones can be explained by intoxication by excessive accumulation of sodium ions which probably induced destruction of cell

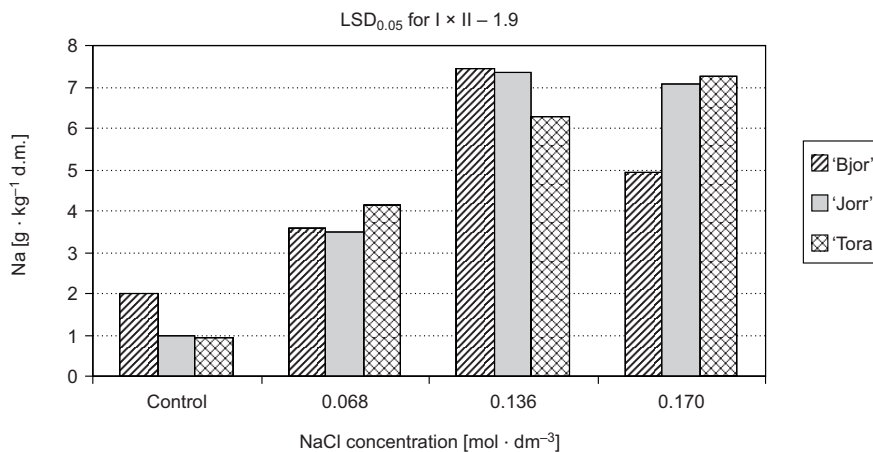


Fig. 1. Content of sodium [g · kg⁻¹ d.m.] in leaves of *Salix viminalis* L. in relation to concentration NaCl in culture

structures and photosynthetic apparatus, which in turn could significantly affect a reduction in water intake.

The increased content of sodium and a small decrease in potassium content in the leaves of *Quercus robur* under NaCl salinity was found by Sehmer et al [10]; in addition, they observed a lower accumulation of some bivalent cations, in particular of Ca²⁺.

The conducted experiment showed that, the salinity did not have any significant effect on the accumulation of potassium in the leaves of the 'Bjor' and 'Jorr' clones (Fig. 2). Only in the 'Tora' clone, a significant increase in the potassium content was found at the concentration of 0.136 mol NaCl · dm⁻³ in the medium (by about 40 %). On the other hand, no significant effect of medium salinity was found on the content of bi-

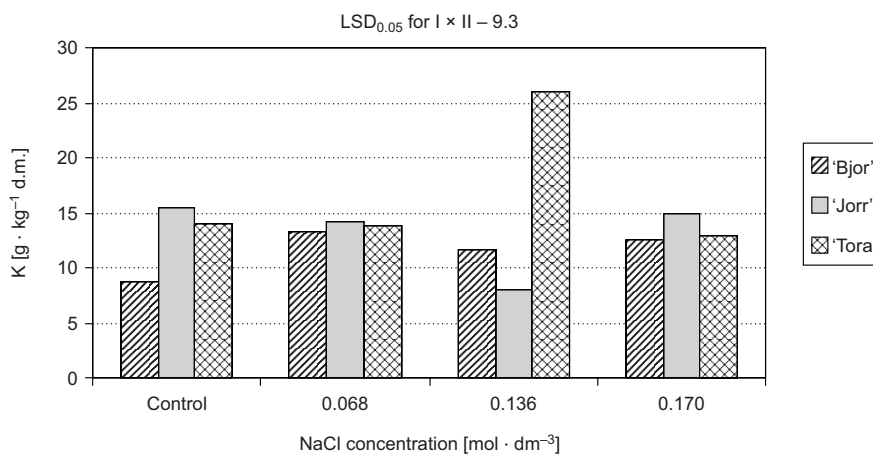


Fig. 2. Content of potassium [g · kg⁻¹ d.m.] in leaves of *Salix viminalis* L. in relation to concentration NaCl in culture

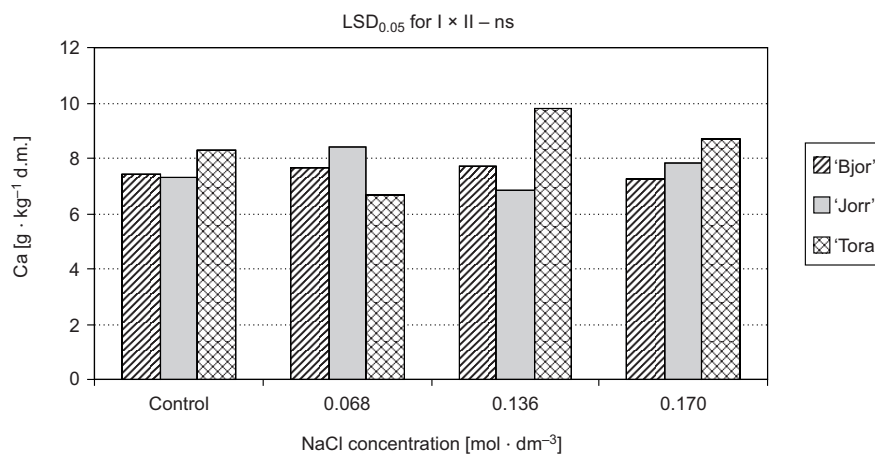


Fig. 3. Content of calcium [g · kg⁻¹ d.m.] in leaves of *Salix viminalis* L. in relation to concentration NaCl in culture

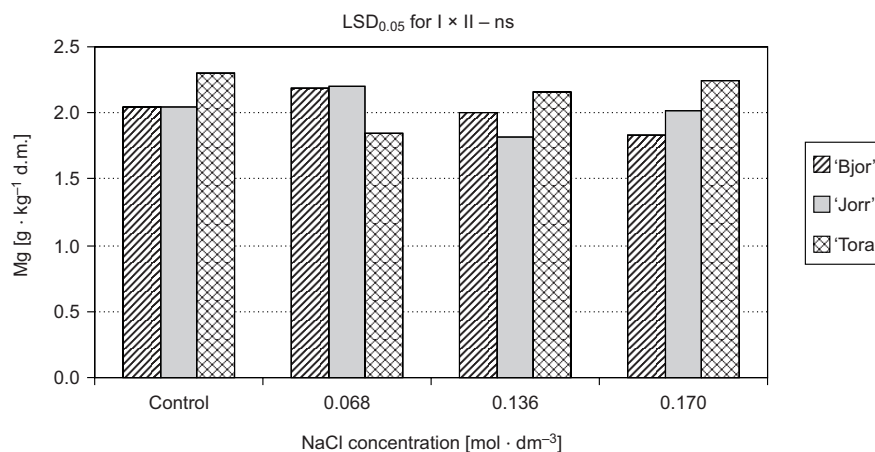


Fig. 4. Content of magnesium [g · kg⁻¹ d.m.] in leaves of *Salix viminalis* L. in relation to concentration NaCl in culture

valent cations, ie calcium and magnesium (Figs. 3 and 4). Wrobel and Gregorczyk [11] showed in their study a significant correlation between calcium and magnesium accumulation in the leaves of the same basket willow clones and soil salinity. However, they applied two times higher doses of NaCl and carried out their experiment under different conditions.

Thus, it appears that the equal level of potassium in the leaf tissues of the 'Bjor' clone, with the limited uptake of excessive quantity of sodium, was decisive for the optimum hydration of leaf tissues at the high concentration of salt in the medium. Additionally, in the leaves of the 'Bjor' clone, out of three clones examined, were observed the most favourable equivalent ion proportions between K:Ca + Mg (0.4 to 0.6), K:Ca

(0.6 to 0.9) and K:Mg (1.3 to 2.1), which were characterised by the smallest fluctuations in the proportion of monovalent to bivalent cations in respective salt concentrations (Table 2). This is exactly them which decide about the optimum hydration of cell plasma and of whole plants.

Table 2

Mean equivalent ion proportions in the leaves of three clones of *Salix viminalis* L.

Clone	Ionic proportions	Concentration of NaCl in culture [mol · dm ⁻³]			
		0.0 (Control)	0.068	0.136	0.170
'Bjor'	K:(Ca + Mg)	0.4	0.6	0.5	0.6
	K:Ca	0.6	0.9	0.8	0.9
	K:Mg	1.3	1.9	1.8	2.1
	K:Na	2.4	2.1	0.9	1,5
	Ca:Mg	2.2	2.1	2.3	2.4
'Jorr'	K:(Ca + Mg)	0.7	0.6	0.4	0.7
	K:Ca	1.1	0.9	0.6	1.0
	K:Mg	2.4	0.5	1.3	2.2
	K:Na	10.0	0.5	0.6	1.2
	Ca:Mg	2.2	2.3	2.3	2.3
'Tora'	K:(Ca + Mg)	0.6	0.8	1.0	0.5
	K:Ca	0.9	1.1	1.3	0.8
	K:Mg	1.9	2.4	3.7	1.7
	K:Na	9.0	2.0	2.4	1.1
	Ca:Mg	2.2	2.2	2.7	2.3

Conclusions

1. Under high NaCl salinity conditions in the medium, the 'Bjor' clone was characterised by more favourable water and ionic balance when compared with the 'Jorr' and 'Tora' clones.

2. From among three basket willow clones examined, only the 'Bjor' clone showed an effective mechanism of decreasing the accumulation of sodium ions in the leaves under the highest salinity of hydroponics.

3. High values of RWC indicator and low values of WSD indicator at the highest concentration of sodium chloride in hydroponics as well as suitable ion proportions, irrespective of the degree of medium salinity, found in the 'Bjor' clone show that it is best adapted to survival under salt stress conditions when compared with the 'Jorr' and 'Tora' clones.

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BILANS WODNY ORAZ JONOWY W LIŚCIACH WIERZBY WICIOWEJ (*Salix viminalis* L.) UPRAWIANEJ W KULTURACH WODNYCH O RÓŻNYM STOPNIU ZASOLENIA

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Abstrakt: W badaniach wykazano ważny wpływ zróżnicowanego stężenia NaCl w pożywce Hoaglanda na bilans wodny i jonowy w liściach trzech klonów *Salix viminalis* L.: 'Bjor', 'Jorr' i 'Tora'. Pomiar wskaźników wodnych RWC i WSD oraz zawartości kationów jednowartościowych (K^+ i Na^+) oraz dwuwartościowych (Ca^{2+} i Mg^{2+}) w liściach klonów wierzby wiciowej pozwalają wnioskować, że w warunkach dużego zasolenia hydroponiki chlorkiem sodu klon 'Bjor' w porównaniu z klonami 'Jorr' i 'Tora' charakteryzował się korzystniejszym bilansem wodnym oraz jonowym, a także skutecznym mechanizmem zmniejszania pobierania jonów sodu w liściach w warunkach największego stężenia soli w podłożu. Wyniki badań wskazują, że klon 'Bjor' jest najlepiej przystosowany do przetrwania w warunkach stresu solnego.

Słowa kluczowe: hydroponika, zasolenie, *Salix viminalis* L., bilans wodny, bilans jonowy