

## STOMATIC CONDUCTANCE AND CHLOROPHYLL CONTENT INDEX AND LEAF AREA OF SOME BEANS LOCAL CULTIVARS FROM NORTH-EAST OF ROMANIA, UNDER SALT STRESS

### DETERMINAREA CONDUCTIVITĂȚII STOMATICE ȘI A CONȚINUTULUI DE CLOROFILĂ A UNOR CULTIVARE LOCALE DE FASOLE, DIN REGIUNEA DE NORD-EST A ROMÂNIEI, SUB INFLUENȚA STRESULUI SALIN

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**Abstract.** Beans is a salt-sensitive species. For this reason, the purpose of the face work was to determine the effect of excess NaCl on the some physiological processes such as photosynthesis and transpiration. Research has focused on dynamics of the chlorophyll content index and foliar stomatic conductivity, in the case of 7 bean genotypes, as an indicator of salt stress tolerance. The biological material was represented by seven bean genotypes, collected from saline soils in the Moldavian region, in 2018 and exposed to salt stress over a 30-day period. They were constantly wetted with saline at a concentration of 100 mM and 200 mM NaCl. Stomatale conductance decrease is a mechanism of resistance to salinity as it prevents water loss from plants. As a response to osmotic component of salt stress to reduce transpiration stomata are partially closed, so we can conclude that genotypes Blăgești 2 shows better tolerance to osmotic stress, compared with other genotypes. After 30 days at 200 mM NaCl, the genotypes Blăgești 2, Blăgești 3, Blăgești 4 and Trușești 2 are superior to the control plants, indicating a good adaptation to intense photosynthetic rhythm. Saline stress influences stomatic conductivite foliar and chlorophyll content, causing significant differences between genotypes.

**Key words:** salinity, beans, chlorophyll, stomata, conductance

**Rezumat.** Fasolea este o specie sensibilă la sare. Din acest motiv, scopul lucrării de față a fost acela de a determina efectul excesului de NaCl asupra unor procese fiziologice, cum ar fi fotosinteza și transpirația. Cercetările s-au axat pe dinamica indicele conținutului de clorofilă și a conductivității stomatice foliare, în cazul a 7 genotipuri de fasole, ca indicator al toleranței la stresul salin. Materialul biologic a fost reprezentat de șapte genotipuri de fasole, colectate din soluri salină în regiunea Moldovei, în anul 2018 și expuse la stresul salin pe o perioadă de 30 de zile. Acestea au fost udate constant cu soluție salină la o concentrație de 100 mM și 200 mM NaCl. Reducerea conductivității stomatale este un mecanism de rezistență la salinitate, deoarece previne pierderea apei din plante. Ca răspuns la componența osmotică a stresului salin pentru reducerea transpirației, stomatele sunt parțial închise, astfel încât putem concluziona că genotipul Blăgești 2 prezintă o toleranță mai bună la stresul osmotic, comparativ cu alte genotipuri. După 30 de zile la tratamentul cu 200

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*mM NaCl, genotipurile Blăgești 2, Blăgești 3, Blăgești 4 și Trușești 2 sunt superioare plantelor control, indicând o bună adaptare la ritmul fotosintetic intens. Astfel, se poate concluziona că stresul salin influențează conținutul conductivității stomatice, dar și conținutul de clorofilă a frunzelor, provocând diferențe semnificative între genotipuri.*

**Cuvinte cheie:** salinitate, fasole, clorofilă, stomată, conductivitate

## INTRODUCTION

Soil salinity it is among the major agricultural problems limiting plant growth and development throughout the world (Adcock *et al.*, 2007; Cantrell and Linderman, 2001). Currently, 20% of the irrigated areas in the world are affected by salinity due to climate change and excessive irrigation. Saline stress is the most devastating abiotic stress, because it affects crop productivity. It diminishes growth, seed germination, photosynthesis and yield (Galani, 2014). Such stress affects agricultural crops by causing an increase in concentration, which leads to a reduction of salts around the root system of the osmotic potential of the water, as it is difficult to absorb water by plants; also induce symptoms of toxicity (ionic effect) (Munns, 2002). Glycophytes, which includes most crop plants, cannot grow in the presence of high salt levels; their growth is inhibited or even completely prevented by NaCl concentrations of 100-200 mM, resulting in plant death (Munns and Termaat, 1986). Salinity can affect plant growth in three ways (James *et al.*, 1982): it can increase the osmotic potential, reducing water availability, respectively the osmotic effect. Munns (1992) concluded that the salts absorbed by plants do not control growth directly, but that they do influence, photosynthesis and/or the activity of specific enzymes.

## MATERIAL AND METHOD

The biological material was represented by seven bean genotypes, collected from saline soils in the Moldavian region, in 2018 and exposed to salt stress over a 30-day period. They were constantly wetted with saline at a concentration of 100 mM and 200 mM NaCl. The bifactorial experience was performed in a randomized three-repeat block experiment. The chlorophyll content of the leaves was determined using the CCM 200 PLUS apparatus. Stomatal conduction was determined using the SC-1-Terra-Preta foliar porometer (Jitareanu *et al.*, 2014). The results were statistically interpreted using the Student test model determined with Microsoft Excel Data Analysis (Oancea, 2007).

## RESULTS AND DISCUSSIONS

**Effect of saline stress on stomatic conductivity.** In most unfavorable conditions, dehydration is the first signal that induces the plant response, consistent with maintaining the status of water in tissues. The condition of keeping the stomata relatively open to plants is the efficient delivery of water to the leaves. In fact, the decrease in the hydraulic conductivity of the leaves is usually caused by the decrease in the degree of hydration associated with the reduction of the stomatic conductivity and, consequently, the reduction of carbon assimilation (Melenciuc, 2010, Sperry *et al.*, 2002, Johnson *et al.*, 2009). The stomatic conductivity foliar to the bean plants in experience

was determined using porometre, and the readings were made at 15 and 30 days after the application of saline treatments.

The stomatic conductivite foliar to 15 days after the application of saline treatments with 100 mM NaCl compared to the witness, was decreasing to three of the populations taken in the study. The genotype with the highest stomatal conductance was Moșna (94.81 mmol/m<sup>2</sup>s), and the Săveni showed the lowest stomatic conductance (40.30 mmol/m<sup>2</sup>s), presenting a reaction to the osmotic component of saline stress, to reduce trash; in this case the stomata were partially closed, as also reported by Galan (2014) (tab.1); that present results confirm for the four populations that the data from the literature, according to which the stomata are partially closed, as a reaction to the osmotic component. Similar results were recorded by Munns and Tester (2008), Covașă (2016), Galani (2014).

Table 1

**The 15 days effect of the saline stress on the foliar stomatal conductance on bean (mmol/m<sup>2</sup>s)**

Genotype	Control	100 mM NaCl	200 mM NaCl
Blăgești 1	81.39	59.06	27.40
Blăgești 2	34.29	49.88	42.44
Blăgești 3	74.51	42.96	37.76
Blăgești 4	38.01	68.18	40.40
Trușești 2	50.42	60.22	80.12
Săveni	52.64	40.30	42.14
Moșna	91.82	94.81	82.37

After 30 days of saline treatment, with regard to stomatic conductivite foliar to the variant treated with 100 mM NaCl, compared to the witness, the genotype Blăgești 2 recorded the highest value (65.34 mmol/m<sup>2</sup>s), instead the Moșna population showed the lowest stomatic conductivite foliar (30.46 mmol/m<sup>2</sup>s) (tab. 2). At the end of the saline treatment, no specific symptoms of ionic stress, chlorosis and necrosis were observed, indicating that the local populations taken in the study are maintained in the first phase of saline stress, making them tolerant to the salin concentrations used.

Table 2

**The 30 days effect of the saline stress on the foliar stomatal conductance on bean (mmol/m<sup>2</sup>s)**

Genotype	Control	100mM NaCl	200 mM NaCl
Blăgești 1	46.54	35.73	24.82
Blăgești 2	62.31	65.34	71.23
Blăgești 3	41.06	41.64	43.45
Blăgești 4	82.02	74.46	63.56
Trușești 2	74.99	46.56	42.33
Săveni	28.64	58.30	61.02
Moșna	36.66	30.46	25.91

Following the statistical analysis (tab. 3) for local bean population subject to saline stress, there are statistically significant differences between genotypes within the same variants, indicating a different behavior to saline stress, depending on the genetic capacity of each genotype. Insignificant differences are highlighted between the populations of different variants, which suggests to us that stomatal conductance recorded values close to the control group. Thus we can conclude that the local populations taken in the study have a good ability to adapt to saline stress.

Table 3

**The variance analysis of the bean genotypes under stress salinity of stomatal conductance (after 30 days)**

Source of variation	SP	GL	MS	F	P-value	F crit	Influence
Genotype	3630.136	5	726.0272	4.766066	0.01734	3.32583	*
Concentration	27.54591	2	13.77296	0.090414	0.91429	4.102821	NS
Error	1523.326	10	152.3326				
Total	5181.008	17					

**Anova Two-Factor:** NS statistical insignificant differences ( $p \geq 0.05$ ); \* Significant statistical differences ( $p \leq 0.05$ ); \*\* Significant distinct statistical differences ( $p \leq 0.01$ ); \*\*\* very significant statistical differences ( $p \leq 0.001$ )

**Effect of saline stress on the chlorophyll content index (CCI).** After 15 days when applying 200 mM treatment, the chlorophyll content was superior to only one population, taken in the study, and lower for the other five populations; the highest increase of the chlorophyll content was found in Blăgești 3 (27.78 CCI), and Trușești 2 recorded the smallest increase (16.26 CCI) (tab.4). The results obtained show an increase of this index in plants exposed to saline stress, indicating that populations with a high index are found in the first phase of saline stress (Munns, 2008).

Table 4

**The 15 days effect of the saline stress on the chlorophyll content index (unit. SPAD)**

Genotype	Control	100 mM NaCl	200 mM NaCl
Blăgești 1	32.15	32.04	26.09
Blăgești 2	14.2	22.73	21.68
Blăgești 3	31.44	33.37	27.78
Blăgești 4	23.91	26.57	21.91
Trușești 2	16.84	15.31	16.26
Săveni	33.52	25.41	26.01
Moșna	24.77	20.69	19.21

From the data presented in tab.5 it can be observed that after 30 days, the chlorophyll content in the leaves presents values which to most bean populations, when exposed to 100 mM NaCl, is inferior to all studied populations ranging from 11.33 to 22.21 CCI, which indicates the passage of genotypes to the second phase of saline stress, the ionic stress. On the other hand, local populations Blăgești 1, Săveni and Moșna show lower values compared to the control variant, which indicates that the second phase of saline stress manifests (Munns and Tester, 2008). At the end of the saline treatment, no specific symptoms of ionic stress, chlorosis and necrosis, were observed, suggesting that the local populations under study remain in the first phase of saline stress, making them tolerant to the saline concentrations used.

Table 5

**The 30 days effect of the saline stress on the chlorophyll content index (unit. SPAD)**

Genotype	Control	100 mM	200 Mm
		NaCl	NaCl
Blăgești 1	15.74	13.87	12.81
Blăgești 2	10.15	11.33	13.75
Blăgești 3	20.98	20.21	22.39
Blăgești 4	18.65	13.54	23.71
Trușești 2	14.02	12.35	15.8
Săveni	26.09	13.63	20.74
Moșna	22.94	14.76	21.28

The statistical analysis of the influence of saline stress on the content of chlorophyll pigments (tab.6) in the local bean populations studied, presents both statistically significant differences between genotypes within the same variants, as well as between genotypes within different variants, indicating that this parameter is significantly influenced by saline stress, and the plants under study show an intense reaction to adapt to this abiotic stress.

Table 6

**The variance analysis of the bean genotypes under stress salinity of the chlorophyll content index (after 30 days)**

Source of variation	SP	GL	MS	F	P-value	F crit	Influence
Genotype	214.7833	5	42.95665	5.102013	0.013924	3.325835	*
Concentration	98.18823	2	49.09412	5.830967	0.020966	4.102821	*
Error	84.1955	10	8.41955				
Total	397.167	17					

**Anova Two-Factor:** NS statistical insignificant differences ( $p \geq 0.05$ ); \* Significant statistical differences ( $p \leq 0.05$ ); \*\* Significant distinct statistical differences ( $p \leq 0.01$ ); \*\*\* Verry significant statistical differences ( $p \leq 0,001$ ).

## CONCLUSIONS

1. Stomatal conductance decrease is a mechanism of resistance to salinity as it prevents water loss from plants. As a response to osmotic component of salt stress to reduce transpiration stomata are partially closed, so we can conclude that genotypes Blăgești 2 shows better tolerance to osmotic stress, compared with other genotypes.

2. After 30 days at 200 mM NaCl, the genotypes Blăgești 2, Blăgești 3, Blăgești 4 and Trușești 2 are superior to the control plants, indicating a good adaptation to intense photosynthetic rhythm.

3. Saline stress influences stomatic conductivite foliar and chlorophylle content, causing significant differences between genotypes.

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