MORPHO-PHYSIOLOGICAL AND PRODUCTIVITY PERFORMANCES OF ESENȚIAL WHEAT VARIETY IN SALINE CONDITIONS

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Abstract. *Romania's significant wheat production capacity, ranking among the top European and global producers, underscores its crucial role in contributing to food security. Addressing the challenge of soil salinization is essential for sustaining agricultural productivity in Romania, and testing wheat varieties for salinity tolerance represents a promising strategy to mitigate the negative impact of this abiotic stress. For this purpose, an experiment was conducted to test the tolerance and adaptability of the Esențial wheat variety to six gradually increasing saline doses under field conditions. The observed trends were unexpectedly indicative of the growth and development parameters, including plant height and spike length, which recorded the best performance at 75 mM NaCl, while the greatest increase in awn length occurred at 45 mM NaCl. Chlorophyll content varied throughout the experiment, initially peaking at 45 mM NaCl but ultimately being highest at 75 mM NaCl. Moreover, productivity parameters, except for water percentage, also demonstrated their peak performance at 75 mM NaCl. The highest water percentage was observed in plats tested with 30 mM NaCl. Additional research should explore the salinity tolerance of different wheat varieties under a range of saline levels to enhance the understanding of their adaptive responses and to pinpoint the most resilient varieties for cultivation in saline-affected areas.*

Keywords: *Leaf greenness, saline dosage, stress adaptation, threshold effect, wheat growth*

INTRODUCTION

Regarding ensuring food security not only at the European level, Romania plays a crucial role through its wheat production (CHIURCIU et al., 2023). In 2023, out of the total area cultivated with major crops, approximately 63% was represented by cereal crops for grain, and another 28% by wheat and rye (INSSE, 2024). According to statistics, wheat and rye production reached approximately 10 million tons in 2023 (INSSE, 2024). Romania, in 2020, was classified among the Group I global wheat producers (GAGIU et al., 2023), and in 2021 was the third largest wheat producer in European Union (CHIURCIU et al., 2023). In the 2021/2022 agricultural year, Romania exported over 6.5 million tons of wheat (VENIG et al., 2023). Given the economic instability caused by the conflict in Ukraine, Romania remains one of Europe's key agricultural producers (MATEOC SÎRB et al., 2023).

Within the country's borders are some of the most fertile soils in Europe (DUMEA COPCEA et al., 2013), a fact that could make them suitable for wheat cultivation. In addition to pedological conditions, climate also plays an important role for wheat crop (CROITORU et al., 2012). However, extreme weather events, such as severe droughts followed by massive floods, create a significant vulnerability for agriculture (COGATO et al., 2019). Another issue that cannot be overlooked is soil salinization. This phenomenon, already affecting certain areas of the country, can have a major impact on agricultural productivity (STANCIU, 2024). In Romania, over 600,000 hectares of land are affected by salinization and alkalization (MIHALACHE et al., 2015). Areas with potential to be affected by salinity are found along the western border, in Dobrogea, southern Moldova, and along the Danube plain (TOTH et al., 2008). Soil salinization reduces agricultural yields (SINGH et al., 2022) by degrading soils through natural salt accumulation, depending on the nature of the parent rock, and through limited freshwater resources (HAYAT et al., 2020). Additionally, excessive irrigation, the lack of efficient drainage systems, and the heavy use of fertilizers contribute to soil salinization after water evaporation (CUEVAS et al., 2019).

The effects of salinization are not limited to soil degradation; they also impact the growth and development processes of plants (SAFDAR et al., 2019). The osmotic stress caused by saline soils induces metabolic changes (HASANUZZAMAN et al., 2013), reflected in altered physiological processes, leading to changes in the morphological parameters of wheat (GUO et al., 2015). The effects of salinity can be observed as early as the germination process, reducing its efficiency (AKBARIMOGHADDAM et al., 2011). Furthermore, high concentrations of salinity affect the critical physiological process of photosynthesis by reducing the content of photosynthetic pigments (AFZAL et al., 2008). All these factors lead to lower yields in key production parameters such as the number of spikes and tillers, ultimately reducing both fresh (SELEIMAN et al., 2022) and dry biomass (ZOU et al., 2016). Moreover, high saline concentrations can hinder the growth and development of wheat plant roots and stems (SELEIMAN et al., 2022).

Given the essential role that wheat crop plays in the country's economy and the need to ensure food security, the issue of soil salinization must not be neglected. In this context, testing wheat varieties from national collections to study salinity tolerance represents a promising area of interest for Romania's agriculture.

The aim of this study was to assess the resistance of the Esențial wheat variety under saline conditions to determine its potential for cultivation in areas affected by this abiotic stress. The objectives of the study were to analyse the effects of salinity on key parameters. The first objective involved monitoring the chlorophyll content of the leaves throughout the experiment, as well as the chlorophyll content of spikes at the end of the experiment. The second objective was to evaluate the effects of saline treatments on the morphological parameters of wheat plants, while the third objective focused on investigating the influence of salinity on key parameters relevant to agricultural production.

MATERIAL AND METHODS

In accordance with the objectives of this study, a mesophytocosm experiment was established in the Agro-botanical Garden (46°45'35.2"N, 23°34'27.7"E) of the University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca campus. The experimental units (mesophytocosms) were filled with soil before sowing wheat seeds of the Esențial variety, obtained from the Agricultural Research and Development Station (ARDS) Turda. Esențial is a common winter wheat (*Triticum aestivum*) variety developed by Agricultural Research and Development Station Suceava in 1988 through the crossbreeding of parental forms and officially registered in 2001 (DOBROTĂ & GONTARIU, 2017). This variety represents a major contribution to Romanian wheat improvement, being classified by ARDS Suceava among the most important varieties between 1996 and 2005. Esențial is a semi-early variety and has been considered adaptable to various environmental conditions (DOBROTĂ, 2016). Salinity stress was introduced at the onset of the experiment by administering a control treatment (0 mM NaCl) alongside five NaCl concentrations in a gradual increasing of 15 mM NaCl. The salt treatments were applied in different concentrations: 15, 30, 45, 60, and 75 mM NaCl, each replicated six times. To prevent contamination of the surrounding soil, experimental pots were isolated with polyethylene mesh. No irrigation was applied, and environmental conditions were left uncontrolled throughout the duration of the experiment. The experiment lasted until the plants entered in the phenological stage senescence, with most parameters being evaluated at the end, except for the relative chlorophyll content of the leaves, which was monitored five times starting from the appearance of the first leaf. The assessments were done at the beginning of every decade of November, at the beginning of April and at the ending of May. This parameter was measured using the MC-

100 Chlorophyll Meter (Apogee Instruments), which provided SPAD units (Soil-Plant Analysis Development) (BARUTÇULAR et al., 2016) readings corresponding to the intensity of green leaf coloration. The same instrument was used to measure the relative chlorophyll content of the spikes at the end of the experiment. Plant height, spike length, and awn length were also measured using the calliper method. Simultaneously, the number of stems (Nr.T), number of spikes (Nr.S), and spike percentage (%Sp) were determined by calculating the ratio of spikes to stems at the end of the experiment. Fresh and dry biomass of the stems (TFW and TDW, respectively) and spikes (SFW and SDW, respectively) were measured using a high-precision analytical balance. The water content in one gram of fresh plant material (%H_2OT/g for stems and %$H_2$0S/g for spikes) was determined by calculating the difference between the fresh and dry biomass, following sample dehydration in an oven at 105°C (MANI et al., 2004) until a constant mass was achieved. After evaluating the parameters of interest, a comprehensive database comprising all replicates was generated, and basic statistical analyses were performed to calculate mean values and standard deviations. Relative chlorophyll content data for both leaves and spikes were represented in figures according to the measurement date using the advanced boxplot function of the free online statistics software, Statistics Kingdom (STATSKINGDOM, 2024). Parameters such as Nr.T, TFW, TDW, %H_2OT/g, Nr.S, SFW, SDW, %$H_2$0S/g, and %Sp were compared to the control plants (0 mM NaCl). The differences from the control (_diffM) were compiled into a table and filtered using conditional formatting to create a diagram. In this way, the range of values for each parameter was color-coded, with the highest values represented in dark red and the lowest performance coded in dark blue.

RESULTS AND DISCUSSIONS

Plant height, spike and awns lengths

The analysis of plant height data reveals an unexpected trend, as salinity is typically associated with adverse effects on plant physiology (TRUȘCĂ et al., 2023). The most pronounced increase in plant height is particularly notable, with the highest value recorded at the highest saline dose (75 mM NaCl), showing an approximate 18% increase compared to the control group (0 mM NaCl) (Table 1).

Table 1.

Esențial wheat variety morphological parameters, plant height, spike length and awns length in cm

Saline doses	Plant height	Spike length	Awns length
0 mM NaCl	39.33±1.56	7.43±0.72	4.70±0.65
15 mM NaCl	45.67±1.54	8.33±0.33	5.00±0.00
30 mM NaCl	38.53±1.92	7.53±0.68	4.37±0.38
45 mM NaCl	39.77±0.95	8.87±0.19	5.23±0.49
60 mM NaCl	40.47±2.59	7.97±0.32	4.77±0.12
75 mM NaCl	47.87±3.19	9.13±0.43	4.70±0.23

This can be associated with the hermetic response to a moderate stress, wherein sub-lethal doses of stressors stimulate growth (JALAL et al., 2021). A similar pattern is observed in plants exposed to 15 mM NaCl, where a reduction of up to 5% from the maximum height was recorded. All other doses, including the control, exhibited height performances within the same value range, with the maximum difference between plant heights varying by no more than 2 cm.

Interestingly, the longest spikes were observed at the highest NaCl concentration, while the shortest spikes were found in the control group, showing a decrease of approximately 19%

from the maximum value (Table 1). The trend of longer spikes under higher salinity, particularly at 75 mM NaCl, suggests that plants might be optimizing their structures to ensure survival and reproduction in adverse environments (SHABALA, 2013). Similarly, reduced spike lengths were observed in plants subjected to 30 mM NaCl, whereas the performance of those exposed to 45 mM NaCl mirrored that of the plants at the highest saline dose. This parameter analysis reveals a trend consistent with that observed in plant height, namely that most plants exposed to saline doses exhibited better performance compared to the control.

Regarding awn length, the highest values were observed at the moderate salt concentration (Figure 1). The application of 45 mM NaCl led to an increase in awn length, whereas a saline environment with a concentration of 30 mM NaCl decreased awn length performance by approximately 7% compared to the control and by 16% compared to the maximum recorded at 45 mM NaCl (Table 1).

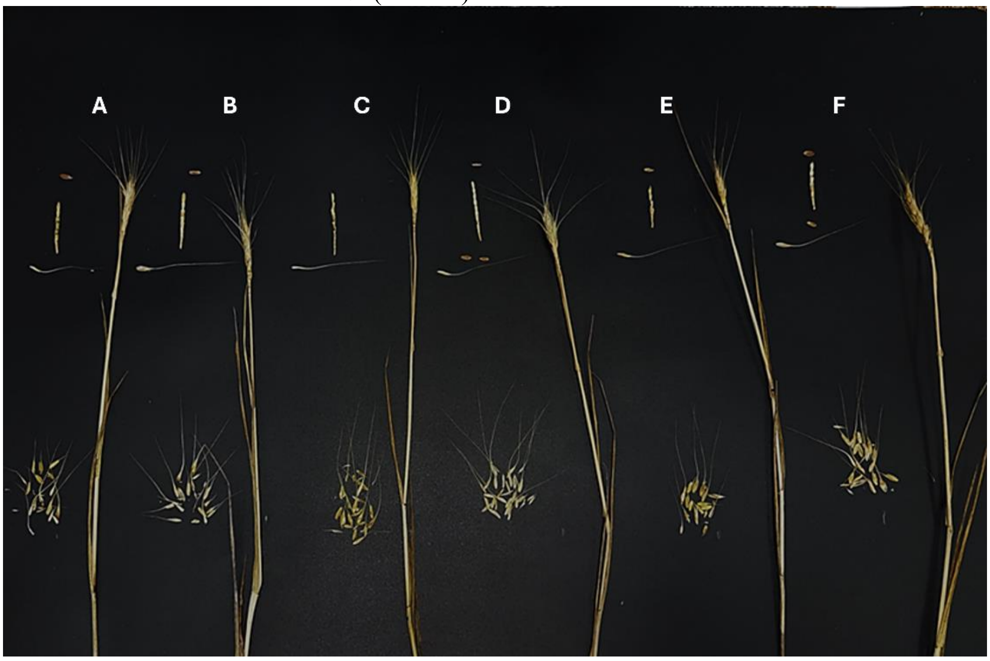

Figure 1. Morphological features of Esențial wheat variety under saline conditions (A:0 mM NaCl, B:15 mM NaCl, C:30 mM NaCl, D:45 mM NaCl, E:60 mM NaCl, F:75 mM NaCl).

On the other hand, the values recorded for the highest saline doses (60 and 75 mM NaCl) and the control were within the same range, with differences of no more than 1.5%. This indicates a dose-dependent response, where intermediate salinity levels promote growth, but deviations from this concentration, either higher or lower, result in reduced performance (GROSZYK & SZECHYŃSKA-HEBDA, 2021).

Leaf and spike chlorophyll content

The relative chlorophyll content, a key indicator of plant health, exhibited dynamic responses to salinity across various growth stages. Initially, the plants had only one unfolded leaf, and due to this early growth stage, chlorophyll levels showed the lowest average values across all measurement periods (Figure 2).

Chlorophyll pigment synthesis was most stimulated by the moderate salt dose (45 mM NaCl). This peak in relative chlorophyll content, expressed in SPAD units, was approximately four times higher than the observed values from the control treatment. The trend observed in the first measurement shows a rise in green colour intensity following the application of saline treatment. Furthermore, a gradual decrease in chlorophyll content was observed as the dose exceeded 45 mM NaCl. Nonetheless, the values recorded at the two highest salt concentrations were still more than double the values observed in the control.

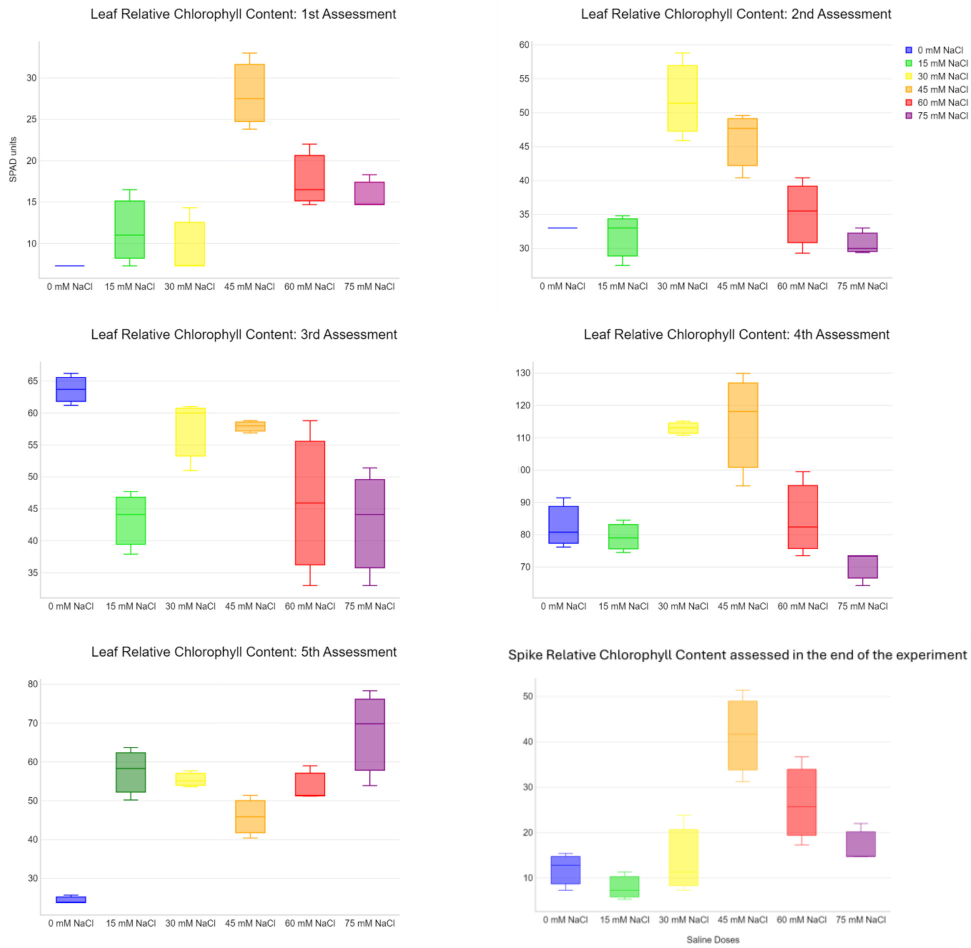

Figure 2. Relative chlorophyll content for leaves, in five assessments, and spike measured at the end of the experiment for Esențial wheat variety.

In the second measurement, saline treatment continued to influence the evolution of relative chlorophyll content. The treated variants (0, 15, 60, and 75 mM NaCl) exhibited values within the same range, with a maximum difference of 5.62 SPAD units (Figure 2). Although the average values did not differ significantly, an important aspect should be noted: between the first and second measurements, the green colour intensity increased fivefold in the control, threefold at 15 mM NaCl, and only approximately twofold at 60 and 75 mM NaCl, compared to the previously recorded values. This highlights that saline stress not only affects chlorophyll content but also prolongs the period during which the parameter can be enhanced as plants continue to develop. On the other hand, the application of 30 and 45 mM NaCl led to an increase in green leaf intensity by approximately 58% and 39%, respectively, compared to the control. This is likely because the 30 mM NaCl dose is sufficiently strong to stimulate chlorophyll production, alleviating the salinity negative effects, but not so concentrated as to disrupt physiological processes. Beyond 30 mM NaCl, higher doses reduced this parameter performance.

The third measurement presented a different distribution of chlorophyll content. The plants from the control treatment doubled their chlorophyll content compared to the previous measurement (Figure 2). Close to the control, the chlorophyll content in SPAD units for the 30 and 45 mM NaCl treatments was grouped with a difference of just 0.5 SPAD units. The plants treated with 15, 60, and 75 mM NaCl, registered around 50% increase compared to their previous values.

The green intensity doubled for plants treated with 30 and 45 mM NaCl, compared to the values recorded in the previous measurement for the same treatments (Figure 2). Relative chlorophyll content in the control reached only three-quarters of the maximum value recorded at 45 mM NaCl. Variants treated with 15 and 60 mM NaCl recorded values similar with the control, while the lowest value was observed under the highest salt concentration.

The final measurement indicated that salinity delayed plant senescence, as chlorophyll levels in treated plants were significantly higher than in the control, which experienced a sharp decline (Figure 2). The recorded values more than doubled for all salinity treatments suggest that the application of saline stress delayed the onset of phenological stages and senescence, being evidence of the higher chlorophyll content.

The relative chlorophyll content of the spikes at the end of the experiment marked a delayed entry into the maturation phase for plants exposed to moderate and high saline doses, indicated by the highest recorded values. While the 15 mM NaCl concentration negatively impacted the parameter, the average values for both the control and 30 mM NaCl were similar (Figure 2). The highest chlorophyll content in spikes was observed in plants stressed with 45 mM NaCl, indicating the longest vegetative period. Beyond this dose, chlorophyll content gradually decreased with increasing NaCl concentration (IBRAHIM et al., 2018). However, the performance at the highest doses remained higher compared to the control. This demonstrates that moderate salinity can extend the plants lifespan, although excessive salinity stress reduces chlorophyll content (RODRIGUEZ COCA et al., 2023).

These results show that moderate salinity, particularly at 45 mM NaCl, enhances chlorophyll content and prolongs vegetative growth, delaying senescence. While higher salinity levels eventually reduce chlorophyll synthesis (SEYMEN et al., 2023), plants still maintain higher chlorophyll levels compared to the control. Overall, the recorded values shown that moderate salinity can improve plant physiological performance, while excessive salinity reduces these benefits and still promotes some physiological adaptation (MUNNS & GILLIHAM, 2015).

Productive performances and water content

Regarding the number of stems, an interesting observation is the performance at the highest saline stress dose, which is similar compared to the control. This suggests the activation of the hormesis phenomenon, where a sufficiently high dose counteracts the effects of saline stress, yet not concentrated enough to negatively impact plant growth and development, therefore the plant develop a defence resistance. This phenomenon is further evidenced by a 24% at the minimum recorded average value at 45 mM NaCl compared to the dose of 60 mM NaCl On the other hand, stem number reduction becomes more pronounced as the dose increases from low to moderate levels (Figure 3). These results suggest that the Esențial wheat variety is suitable for cultivation at concentrations higher than 45 mM NaCl but lower than 75 mM NaCl, for optimal stem production, making it a viable option for biomass production.

For stem fresh biomass, a similar trend of stem number is observed, with a gradual decrease in average values as saline doses increase up to 45 mM NaCl, followed by two successive increases. However, these increasing values remain lower than the control, by approximately 49% (60 mM NaCl) and 6% (75 mM NaCl), respectively.

	M	15 mM NaCl	30 Mm NaCl	45 mM NaCl	60 mM NaCl	75 mM NaCl
Nr.T	30.00	29.00	29.00	22.00	29.00	30.00
Nr T_difM		-1.00	-1.00	-8.00	-1.00	0.00
TFW	6.85	6.00	5.76	3.41	3.48	6.41
TFW_difM		-0.85	-1.09	-3.44	-3.37	-0.44
TDW	6.15	5.40	5.04	3.12	3.27	5.71
TDW_difM		-0.75	-1.11	-3.03	-2.88	-0.44
%H_2OT/g	10.22	10.00	12.50	8.50	6.03	10.92
%H_2OT/g_difM		-0.22	2.28	-1.71	-4.18	0.70
Nr.S	21.00	25.00	24.00	13.00	22.00	26.00
Nr.S_difM		4.00	3.00	-8.00	1.00	5.00
SFW	0.87	1.62	1.52	0.84	1.11	1.70
SFW_difM		0.75	0.65	-0.03	0.24	0.83
SDW	0.76	1.45	1.33	0.75	1.00	1.52
SDW_difM		0.69	0.57	-0.01	0.24	0.76
%H_2OS/g	12.64	10.49	12.50	10.71	9.91	10.59
%H_2OS/g_difM		-2.15	-0.14	-1.93	-2.73	-2.06
%Sp	70.00	86.21	82.76	59.09	75.86	86.67
%Sp_difM		16.21	12.76	-10.91	5.86	16.67

Figure 3. Esențial wheat variety productivity parameters under six salinity doses: mean values (number of tillers (Nr.T), tillers fresh weight (TFW), tillers dry weight (TDW), water percentage in one gram of fresh tillers (%H_2OT/g), spikes number (Nr.S), spike fresh weight (SFW), spike dry weight (SDW), water percentage in one gram of fresh spikes (%$H_2$0S/g) and spike percentage (%Sp) and difference compared with control (_difM)

The trend suggests that doses of 30, 45, and 60 mM NaCl visibly affect fresh biomass registering a decrease under salinity stress (Kausar & Gull, 2014). It can also be stated that, although the number of stems did not change with the highest saline dose, the fresh biomass of those stems decreased, highlighting the impact of saline stress. This can be associated with basic physiological processes connected with osmosis at plant cellular level (Loutfy et al., 2012).

Clearly, the analysis of data reveals a similar trend in stem dry biomass as seen in fresh biomass. In this way, dry biomass decreases with increasing saline doses up to 45 mM NaCl, where the control value is nearly twice lower. A slight increase is then observed with the 60 mM NaCl dose, followed by another visible increase at the highest saline dose. At this concentration, the trend shows a counteracting effect, with the biomass only 7% lower compared to the control.

The percentage of water content in one gram of stems shows a distinct data distribution compared to the trends observed in stem number and fresh/dry biomass. The concentration of 30 mM NaCl stimulated the plants to retain the highest water content, with an increase of approximately 22% compared to the control. Meanwhile, doses of 45 and 60 mM NaCl reduced the water content of plants by approximately 17% and 41%, respectively, compared to the control. At the highest applied saline dose, the phenomenon of hormesis is again observed with a value of 7% higher than the control.

Agricultural production is directly dependent on the number of spikes produced, and the results indicate an interesting trend. Saline stress application stimulated spike growth by approximately 19% at 15 mM NaCl and 24% at 75 mM NaCl. The observed trend suggests a decrease in values as saline doses increase, with higher values than the control up to 45 mM NaCl, where spike number drops dramatically by about 38% compared to the value recorded at 0 mM NaCl. High doses of 60 and 75 mM NaCl stimulate spike formation, as seen in the continuous increase in values.

The analysis of fresh spike biomass data shows a trend similar to that observed for spike number, with nearly double the parameter value at both 15 mM NaCl and the highest saline dose. Additionally, fresh spike biomass does not decrease drastically with the application of any saline dose. This suggests that the Esențial wheat variety is tolerant to the applied doses and highlights the stimulating effect of the highest saline dose on this parameter's performance **(**JALAL et al., 2021**).**

A similar pattern is observed for dry spike biomass as for fresh spike biomass. Water content in the spikes decreases in saline environments, with the minimum observed at 60 mM NaCl, a reduction of approximately 22% compared to the control. The parameter also decreases with the application of 15, 45, and 75 mM NaCl, but the results for these doses fall within the same range, with a maximum difference of 0.22 g. Moreover, water content is not significantly affected by plant growth in a saline environment with 30 mM NaCl.

The spike percentage decreases by approximately 16% in the treatment with 45 mM NaCl. On the other hand, the application of other doses increases the percentage among tested plants, rising by 23% and 24% among those exposed to 15 and 75 mM NaCl, respectively.

The data indicates that the Esențial wheat variety demonstrates a nuanced response to varying saline doses. This fact reflects the degree of tolerance of the genotype to abiotic stressors. The physiological responses of plants to salinity stress depend on a combination of the applied dosage (SIHMAR et al., 2024), pedoclimatic conditions, and the resistance genes possessed by each individual plant (AYCAN et al., 2024). At the highest saline stress (75 mM NaCl), hormesis is evident, where plant performance, including stem number and spike growth increases. The wheat variety performs best at both 15 mM and 75 mM NaCl, suggesting it is relatively salt-tolerant and capable of compensating the saline stress at higher concentrations. For optimal biomass production under abiotic stress, salinity levels above 45 mM but below 75 mM appear most favourable.

CONCLUSIONS

Plant growth and development in height and spike length recorded the best performance at 75 mM NaCl. The concentration of 45 mM NaCl proved to be the most favourable for increasing the length of the awns.

Chlorophyll content registered the lowest performance both at the beginning and the end of the experiment in the control treatment. However, the other notable deficiencies observed throughout the experiment occurred in plants stressed with 75 mM NaCl.

Initially, the chlorophyll content was highest in plants stressed with 45 mM NaCl. Throughout the experiment, the top values fluctuated between 0 and 45 mM NaCl, but by the end, the highest chlorophyll content was observed in plants treated stressed with the highest dose of 75 mM NaCl.

The productivity parameters analysed, apart from water percentage, exhibited the best performance at 75 mM NaCl. Conversely, the lowest biomass values were observed in plants within 45 mM NaCl treatment.

The highest percentage of accumulated water was found in plants subjected to 30 mM NaCl treatment. However, plants grown in the saline environment of 60 mM NaCl exhibited a defective water regime.

Future studies should focus on evaluating the salinity tolerance of multiple wheat varieties across a range of saline doses to gain a comprehensive understanding of their adaptive capacities and to identify the most resilient cultivars for cultivation in saline-affected areas.

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