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Analysis of the germination of two varieties of durum wheat (*Triticum turgidum L.var.durum*) as a function of salinity: *in vitro* approach

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Abstract

The tolerance of two durum wheat varieties was tested under different levels of salinity using an experimental approach in the laboratory. The results show that the Vitron variety performs significantly better than the Simeto variety despite the fact that the tolerance threshold of both varieties to NaCl is lower than 2 gr/ l. The germination capacity of the Simeto variety was significantly lower than that of the Vitron variety which germinated in a relatively average time of 4 days and recorded the highest germination rates, 73% for seeds watered with distilled water, 53% at 2g/l , and 33% at 5g/l of NaCl , a significant decrease in coleoptile and root length was recorded for both varieties, the Simeto variety recorded significantly higher levels of proline than the Vitron variety, 0.346 µg/ml *vs* 0.280 µg/ml under 2 g/l , respectively

Keywords: Durum wheat, salt stress, NaCl, germination, proline, tolerance.

1. Introduction

Wheat is among the cereals that occupies a primary place in the agricultural system, with a world production of 783.8 million tons [1], Algeria is ranked among the lowest producing countries in the world with an annual production of 3.3 million tons during the last five years [2] while the country has consumed 11.37 million tons between 2020 and 2021 according to a report co-produced by the Global Agricultural information [3].

Currently, Algeria wishes to move towards an intelligent and resilient agriculture to climate change, such as the stimulation and specialization of durum wheat production which is more favorable to the nature of Algerian farmland [4]. One way to increase production would be to adapt crops to saline soils or land irrigated with brackish water [5]. Salinity is one of the main abiotic stresses limiting crop growth [6].

In the world, soil salinization is becoming a major agricultural problem, 9% of the world's land surface is affected by salinity and more than two thirds of its saline soils are found in these arid and semi-arid climatic zones [7].

The negative effect of high salinity can be observed at the whole plant level such as plant death and/or decreased productivity [8]. Studying seed behavior to salinity is an indicator of plant salinity tolerances for later stages of seedling development and growth [9]. A good understanding of the factors of early seed germination allows a better classification of genotypes; a late development favours the accumulation of toxic ions leading to the death of plants before the end of their development cycles [10].

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The objective of this work is to evaluate the salinity tolerance of two durum wheat varieties Simeto and Vitron, in order to determine the best environment for seed germination. As well as conducting examinations of biometric characteristics and proline synthesis to better understand the behavior of these two varieties in the face of high NaCl concentrations.

2. Material and methods

2.1. Material

The seeds used in the germination tests were taken from the collection of the Institut National of Soils of El Matmar, Relizane, Algeria. The study was carried out on two durum wheat varieties Simeto and Vitron.

2.2 Methods

The seeds are disinfected and sown in petri dishes whose interior is lined with filter paper, thereafter deposited in a germoir at 25 °C, thereafter, the saline stress is applied to the seeds after immersion at two concentrations of NaCl: (50 meq / l and 100 meq / l) or (2.92g / l and 5.85 g / l) respectively.

In each petri dish containing ten seeds, 10 ml of distilled water were poured for the control seeds and 10 ml of saline solution for the salinity-treated seeds, each treatment is repeated three times. As soon as the tip of the radicle appeared through the husks, we proceeded to count the germinated seeds regularly. When the germination rate stabilized, we completed our observations to begin the second part of the experiment.

2.2.1. Germination parameters

a. Precocity of germination. This parameter was determined when we observe the first germinated seeds , in this case, the precocity of germination was expressed by the rate of the first germinated seeds corresponding to the time interval between the sowing of the seeds and the first germinated seeds [11].

b. Germination time. Germination time is the time (in days) allowed between the first germinated seeds and the end of germination.

c. Germination capacity (germination rate). The germination rate was calculated according to the following equation: % germination= (Ni/Nt)*100

Ni: number of germinated seeds.
Nt: total number of seeds tested.

2.2.2. Study of the biometric parameters. The elongation of the roots and coleoptiles are evaluated by measuring the length after one week of treatment.

2.2.3. Extraction and determination of proline. Proline was determined by the Monneveux and Nemmar method [12], the determination was carried out on young seedlings of 15 days

2.2.4. Statistical analysis. The results are expressed as means and their standard error (X±ES). The data were statistically analyzed using Microsoft Excel version 7.0 software. In all cases, a value of $p<0.05$ was considered significant with : $* P \leq 0.05$; $** : P \leq 0.01$

3. Results and discussion

3.1. Germination parameters

3.1.1. Early germination. The seeds of both varieties germinated from the first day but with different rates (Table 1). The earliest seeds to germinate are those treated with distilled water (DW) for the Virton variety (70%), while lower rates of 47% and 30% are recorded for the treatments with 50 and 100 meq/l of NaCl respectively.

Table 1. Germination earliness (%) of wheat seeds under NaCl salt treatment

Variety	Control	50 meq/l	100 meq/l
Vitron	70 ±1	47 ±1.52**	30±1**
Simeto	57 ±2.08 **	37 ±0.57**	0**

The variety Simeto records lower germination rates, which do not exceed 57% for control seeds, 37% for seeds treated with 50 meq/l NaCl and no germination is recorded with 100 meq/l NaCl, the different salt concentrations have a highly significant effect on the earliness of germination of both varieties Vitron and Simeto. Soil and irrigation water salinity delays the germination initiation process [13] and may inhibit seed germination due to an osmotic and toxic effect [14].

Several similar results on the behavior of seeds towards salt supply have been observed in several wheat varieties [15, 16, 17].

3.1.2. Germination time

The variation in germination time and germination capacity allows the selection of species according to their tolerance and/or sensitivity to salt at germination [18]. For the two varieties studied, seed germination starts on the first day after sowing with varying rates (Figure 1). The control seeds of the variety Vitron recorded the highest rates from the first day 70% and stabilized on the second day to reach 73%. Seeds receiving 50 meq/l of NaCl started germinating at low levels of 47% and 50%, reaching 53% on day 4. As soon as the saline concentration doubles, the duration remains the same with rates reduced by 30%, to stabilize from the last day at 33%.

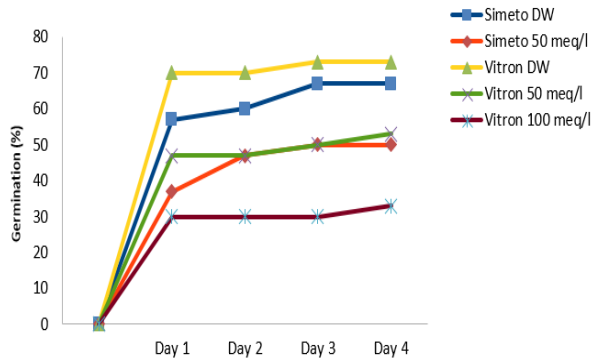

Figure 1 . Germination duration (%) of durum wheat seeds of the two varieties Simeto and Vitron under NaCl.

For the variety Simeto, the control seeds record the highest rates with 57% on the first day to reach 67% on the last day. The seeds treated with saline solution at 50 meq/l record lower rates from the first day with (37%), then a rate of (47%), to reach a rate close to the final rate of control seeds (50%).

3.1.3. Germination capacity (germination rate)

The variety Vitron records a high rate for seeds watered with distilled water 73%, this rate decreases to 53% under 50 meq/l of NaCl and to 33% under 100 meq/ l .

The Simeto variety has lower rates than Vitron, for seeds watered with distilled water the rate is 67%, this rate decreases to 50% for seeds treated with 50 meq.l-1, and to 0% as soon as the NaCl concentration doubles (100 meq/l).

The difference in germination rates of the seeds of the variety Simeto and Vitron, is significant compared to the control for the seeds treated with the NaCl saline solution at 50 and 100 meq/l Nevertheless, this difference is highly significant in the seeds of the variety Vitron which records higher rates than Simeto (table 2).

Studies on similar varieties confirm that salt has a depressive effect on the germination capacity of these seeds [15, 16], the decrease in germination capacity under the effect of salt corresponds to an increase in external osmotic pressure which affects the rate of absorption of water by the seeds [19] and causes significant damage of the cell membrane [20].

Table 2. Germination capacity (%) of durum wheat seeds of both varieties Simeto and Vitron under NaCl.

Variety	Control	50 meq/l	100 meq/l
Vitron	73 ± 1,15	53 ±2,08*	33 ±1,58**
Simeto	67 ±1,15*	50 ±0.57*	0**

Table 3. Changes in coleoptile height and root length of the two durum wheat varieties treated with NaCl.

Variety	Seedlings	Control	50 meq/l	100 meq/l
Simeto	Coleoptiles	11,1 ±0.5	10 ±0.43	0**
	Roots	17,5 ±0.76	10,2 ±0.33**	0**
Vitron	Coleoptiles	9,9 ±1.20	8,6 ±0.45*	5,2 ±0.52**
	Roots	18,3 ±1.22	10,2 ±1.02 **	6,6 ±0.23**

Table 5. Proline content of young seedlings of the two durum wheat varieties treated with NaCl.

Variety	Control	50 meq/l	100 meq/l
Vitron	0,164	0,280 *	0,411**
Simeto	0,220	0,346 *	0**

The accumulation of salts also causes the disruption of enzymes involved in physiological germinative mechanisms such as amylases and peroxidases, which prevents the lifting of embryo dormancy and leads to a decrease in germinative power [21].

3.2. Biometric parameters

The results of the seeds of the two varieties show that after 15 days of sowing, the average length of the radicles is greater than that of the coleoptiles, whether it is for the control seeds or those treated with the saline solution.

There was a significant decrease in coleoptile and root length growth in seeds treated with 50 and 100 meq.l-1 NaCl for both varieties.

Our results are in agreement with the research of [22, 23] on some durum wheat varieties and show that coleoptile length is shorter than roots and that salt causes a decrease in root length.

This decrease is probably due to the absorption of sodium which has toxic effects on seed germination [24], mainly the displacement of Ca^{+2} by Na^{+} in the cell wall, which could disrupt its synthesis and consequently the growth of radicles. Root development can be considered as one of the indicators of resistance to salt stress [25].

3.3. Proline contents

Proline is considered a bio-marker of stress, and helps maintain many physiological functions [26], it has been shown that its synthesis with other organic solutes is overexpressed under salt stress, its accumulation improves the tolerance of plants to salt stress [27], and allows plants to tolerate the lack of water by a decrease in osmotic potential [28].

These studies are consistent with our results, which show that proline accumulates more when the culture medium is enriched with NaCl, for both Simeto and Vitron. The proline content increases significantly from 0.22 µg/ml and 0.164 µg/ ml respectively for the control Simeto and Vitron seeds to 0.346 µg/ ml and 0.280 µg/ ml with a treatment of 50 meq/ l , and to 0.411 µg / ml for Vitron seeds treated with 100 meq /l .

It should be noted that the Simeto variety accumulates more proline compared to the Vitron variety.

4. Conclusion

Our results confirm that the variety Vitron presents an earliness of germination and a rate of germination more important than the variety Simeto. The germination kinetics is in a relatively average time (4 days), on the other hand at high concentrations of NaCl (100 meq/l) no germination is recorded for the variety Simeto. In addition, the presence of NaCl in the culture medium leads to a significant decrease in the length of coleoptiles and roots. The Vitron variety shows a more developed growth compared to the Simeto variety under the different treatments applied. Furthermore, the saline concentrations cause a linear increase in proline levels, especially for the Simeto variety. This suggests that Vitron is more tolerant to salinity than Simeto.

Finally, these results confirm that NaCl is a significant constraint to durum wheat seed germination, and the use of varieties tolerant to soil and irrigation water salinity has become imperative.

Compliance with Ethics Requirements: Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human or animal subjects (if exist) respect the specific regulation and standards.

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