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Evaluation of Durum Wheat Genotypes at Germination Stage under Salinity Stress †

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Abstract: The aim of this work was to investigate several durum wheat genotypes (eleven landraces, two old varieties and seven modern cultivars) for salt tolerance at germination and early growth stages. Seeds were tested under two different concentrations of NaCl solution (50 mM and 100 mM) and a control (T) with distilled water. Experiments were laid out in a two-factorial design with three replications. Two-way ANOVA was performed and means were compared with Duncan's Multiple Range test. Seven parameters were measured under laboratory conditions: germination, mean germination time, shoot length, root length, root number, shoot dry matter and root dry matter. Rusticano, with the highest value of root number (5.44), was statistically different from all other genotypes. Timilia reste bianche, Timilia reste nere, Ciciredda and Cappelli are highlighted for the best root length performance at 100 mM.

Keywords: durum wheat; Sicilian landraces; germination traits; salt stress

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1. Introduction

Salinity is one of the most severe abiotic stress factors affecting plant growth and agricultural production worldwide. It affects almost 1 billion ha worldwide, globally representing about 7% of the continental extent of the earth [1]. Agricultural crops exhibit a wide spectrum of responses under salt stress. Salinity affects almost all aspects of plant development including germination, vegetative growth and reproductive development. It strongly inhibits seed germination through osmotic stress, ion-specific effects and oxidative stress. Sicily, with its variable pedoclimatic conditions, is an important source of agro-biodiversity. In this context, over recent centuries, farmers have made a continuous selection that has led to the creation of numerous landraces.

Landraces are named and maintained by traditional farmers to meet their social, economic, cultural and environmental needs. Durum wheat [(*Triticum turgidum* L. subsp. *durum* (desf.) Husn.] is a typical Sicilian crop with a cultivated area of about 270,000 ha, although on small areas (about 5000 ha), in the last decade, the cultivation of landraces has been reintroduced [2]. Landraces, which have arisen through a combination of natural selection and the selection performed by farmers, usually have a broader genetic base and can therefore provide valuable characteristics important for breeding. To date, 24 Sicilian durum wheat landraces are listed in the national register of varieties (https://www.sian.it/portale-sian/home.jsp consultation in 22 July 2023). Information on the genotypic tolerance to salt stress during the germination process is lacking in Sicilian durum wheat landraces. Limited literature work has been documented so far on

these genotypes, so knowing their behavior can help breeding programs in the selection of salt-tolerant varieties to achieve optimum wheat growth under saline conditions. The aim of this work was to investigate several durum wheat genotypes (eleven landraces, two old varieties and seven modern cultivars) for salt tolerance at germination and early growth stages.

2. Materials and Methods

Laboratory experiments were conducted to evaluate the germination traits of 20 durum wheat genotypes at the Council for Research in Agriculture and Economics—Research Centre for Plant protection and Certification Palermo, Italy in 2023. The seeds of all the genotypes tested were obtained in our experimental station during the growing season 2021/2022 and stored at 5 °C with 30% relative humidity.

Seven modern varieties of durum wheat (Ciclope, Duilio, K-26, Mongibello, Quadrato, Rusticano and Simeto), two old varieties (Capeiti 8 and Trinakria) and eleven landraces (Cappelli, Castiglione glabro, Ciciredda, Francesa, Perciasacchi, Russello, Russello Ibleo, Scorsonera, Timilia reste bianche, Timilia reste nere and Urria) were assessed. Seeds were tested under 2 different concentrations of NaCl solution (50 mM and 100 mM) and a control (T) with distilled water. Seeds were surface-sterilized in 5% sodium hypochlorite solution for 3 min and washed thoroughly under tap water for 10 min and at the end with distilled water. To avoid water losses, the edges of Petri dishes were tightly sealed with an impermeable colorless parafilm. Seeds were allowed to germinate at 20 °C in the dark.

To each Petri dish, sterilized in an oven at 120 °C for 2 h, 12 mL of solution was added to keep the filter paper uniformly soaked without flooding. Fifty randomly selected seeds for each variety were placed in Petri dishes with a 13.5 cm diameter (one replication was made up of 2 Petri dishes containing 25 seeds each) on a double layer of Whatman filter paper No. 1 with the crease facing down and kept in a thermostatic cabinet (KW Scientific Equipment model WRS 85). Shoot and root lengths and fresh and dry weights were recorded after 7 days. Data for shoot and root lengths were obtained from 50 seedlings in each replication.

The seedlings' fresh and dry weights were taken with the help of a digital balance (Mettler Toledo PR503 Delta Range); dry weight was measured by placing them at 80 °C in a hot air oven for 24 h until a constant weight was observed.

Experiments were laid out in a two-factorial design with three replications using a complete randomized design (CRD). A two-way ANOVA was performed and means were compared with Duncan's Multiple Range test at a 5% level of probability. The assumption of normality and homoscedasticity was verified with Shapiro–Wilk and Levene's tests, respectively.

Seven parameters were measured under laboratory conditions: germination, mean germination time (MGT), shoot length, root length, root number, shoot dry matter and root dry matter.

Germination percentages were recorded daily up to the 7th day using radicle extrusion ($\geq$2 mm long) as a criterion. A seed was considered to show abnormal germination if shoot growth occurred in the absence of radicle extension. They are expressed as the ratio of germinated seeds on the 7th day to the total number of seeds using the following formula:

$$G = (c/a) \times 100$$

where a = total number of seeds, c = number of germinated seeds on the 7th day.

Mean germination time (MGT, days): This parameter is determined according to the following formula [3]:

$$MGT = \sum (ni \times di)/\Sigma\, b$$

where n is the number of seeds germinated on day i, d is the incubation period in days, and b is the total number of seeds germinated upon treatment.

To compute shoot and root dry matter, the following formula was used:

$$\text{Dry matter (\%)} = (\text{Dry weight/fresh weight}) \times 100$$

3. Results

Genotype, concentration and their interaction showed significant differences in many of the parameters studied (Table 1).

Table 1. Factorial analysis of variance.

Source of Variation	df	Germination (%)	MGT	Shoot Length (cm)	Root Length (cm)	Root Number (n°)	Shoot Dry Matter (%)	Root Dry Matter (%)
Genotype	19	***	***	***	***	***	***	**
Concentration	2	**	**	***	***	***	***	n.s.
Genotype × Concentration	38	***	n.s.	n.s.	*	n.s.	n.s.	n.s.

*** significantly different at $p < 0.001$; ** significantly different at $p < 0.01$; * significantly different at $p < 0.5$; n.s. not significantly different.

As shown in Table 2, Rusticano, Ciclope, Simeto and Mongibello are notable for the significantly shorter mean germination times (ranging from 2.15 to 2.20 days) compared to Cappelli, Francesa, Perciasacchi and Russello Ibleo (ranging from 2.43 to 2.68 days). In general, all the landraces were reported to have better shoot lengths than modern varieties; the top values are for Ciciredda, Scorsonera, Timilia reste nere and Timilia reste bianche with 7.95, 7.85, 7.76 and 7.69 cm, respectively, values significantly higher than all modern varieties.

Table 2. Influence of genotype on some parameters studied.

Genotypes	MGT		Shoot Lenght (cm)		Root Number (n°)		Shoot Dry Matter (%)		Root Dry Matter (%)	
Rusticano	2.15	f	6.03	efg	5.44	a	9.99	bc	13.17	abc
K-26	2.26	def	6.68	cde	4.83	cde	10.19	b	12.86	abcd
Capeiti 8	2.55	bc	7.61	abc	4.62	ef	9.28	cdef	13.29	ab
Ciclope	2.19	ef	5.66	fg	4.80	cde	10.52	ab	11.65	bcde
Cappelli	2.43	cd	7.09	abcd	4.12	h	8.57	fg	10.46	e
Scorsonera	2.29	def	7.85	a	4.46	fg	9.37	cde	12.89	abcd
Mongibello	2.20	ef	6.65	cde	5.13	b	9.89	bcd	10.92	cde
Quadrato	2.31	def	6.46	def	5.07	bc	10.42	ab	13.41	ab
Francesa	2.43	cd	7.13	abcd	4.66	def	9.15	defg	12.90	abcd
Ciciredda	2.24	def	7.95	a	4.22	gh	8.52	fg	11.63	bcde
Castiglione glabro	2.39	cd	7.21	abcd	4.62	ef	8.66	efg	12.85	abcd
Russello	2.33	def	7.32	abcd	4.44	fg	8.48	g	12.43	abcde
Russello Ibleo	2.68	ab	7.60	abc	4.48	fg	9.77	bcd	11.34	bcde
Duilio	2.35	de	6.04	efg	4.94	bcd	9.83	bcd	12.33	abcde
Timila reste nere	2.39	cd	7.76	ab	3.96	h	8.64	efg	10.47	e
Timilia reste bianche	2.39	cd	7.69	ab	4.07	h	8.49	g	11.41	bcde
Perciasacchi	2.65	ab	6.82	bcde	4.95	bc	8.98	efg	13.14	abcd
Simeto	2.19	ef	5.34	g	4.42	fg	11.02	a	14.00	a
Urria	2.29	def	7.05	abcd	4.42	fg	9.00	efg	11.44	bcde
Trinakria	2.74	a	6.49	def	4.57	ef	8.91	efg	10.85	de

In each column, means followed by the same letter are not statistically different according to Duncan's Multiple Range test at the 5% probability level.

Rusticano showed the highest value of root number (5.44), statistically different from all other genotypes; however, Timilia reste nere, Timilia reste bianche, Cappelli, Ciciredda were characterized by the lowest values.

In general, modern varieties showed significantly higher shoot dry matter values than landraces except for Francesa, Scorsonera and Russello Ibleo.

A rise in NaCl concentration determined an increase in the mean germination time and shoot dry matter; mean germination time passed from 2.31 days (control) to 2.38 days (50 mM) and 2.43 days (100 mM), while shoot dry matter varied from 8.92 (control) to 10.04 (100 mM). Increasing the concentration of NaCl in the solution led to a significant reduction in shoot length from 7.73 cm (control) to 5.67 cm (100 mM). The root number was higher at 50 mM (Figure 1).

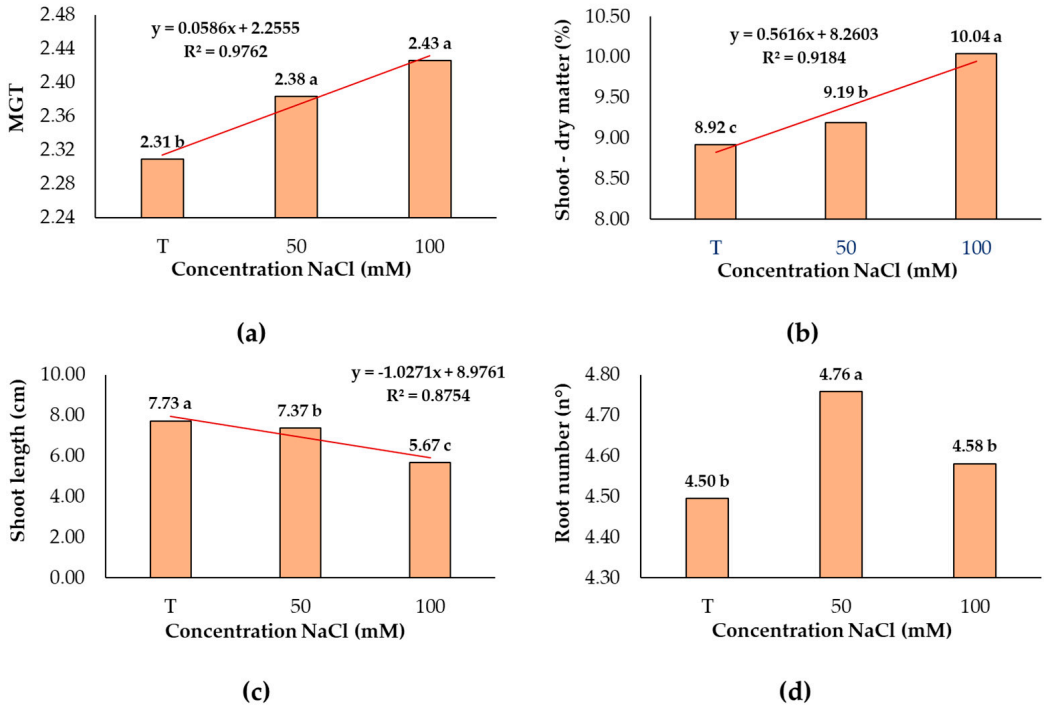

Figure 1. Influence of concentration on (**a**) Mean germination time (MGT); (**b**) Shoot dry matter (%); (**c**) Shoot length (cm); (**d**) Root number. Means followed by the same letter are not statistically different according to Duncan's Multiple Range test at the 5% probability level.

Remarkable germination values, at 100 mM, were obtained by Timilia reste nere (98.67%), Ciclope (98.00%), Mongibello (96.67%) and Timilia reste bianche (96%); in particular, Timilia reste nere showed the highest value, statistically different from the two modern varieties Duilio and Quadrato; Trinakria, Scorzonera, Russello and Russello Ibleo reported the lowest ones, statistically different from all other genotypes.

At 50 mM, the landraces Cappelli, Urria, Timilia reste bianche and Timilia Reste nere showed good performance of germination, while Trinakria, with the lowest value, was statistically different from the other varieties tested.

The root length best performances at 100 mM were achieved by Timilia reste bianche (7.88 cm), Timilia reste nere (6.84 cm), Ciciredda (6.75 cm) and Cappelli (6.54 cm); Perciasacchi, Russello Ibleo and Duilio were statistically different to Ciciredda, Timilia reste bianche and Timilia reste nere (Table 3).

Table 3. Influence of the concentration × genotype interaction on some parameters studied.

Concentration × Genotype		Germination %		Root Lenght (cm)	
0 mM	Capeiti 8	97.33	abc	6.89	fghijklmnopqrstuv
	Castiglione glabro	97.33	abc	7.83	cdefghijk
	Ciciredda	95.33	abcdef	7.90	cdefghij
	Ciclope	92.67	abcdefghi	9.22	abc
	Duilio	94.00	abcdefghij	7.72	defghijkl
	Francesa	88.67	fghijkl	8.12	cdefgh
	K-26	90.00	defghijk	6.99	defghijklmnopqrst
	Mongibello	93.33	abcdefghi	7.91	cdefghij
	Perciasacchi	89.33	efghijk	7.05	defghijklmnopqrs
	Quadrato	94.33	abcdefgh	6.94	efghijklmnopqrstu
	Russello	88.00	ghijkl	8.45	bcd
	Russello Ibleo	83.33	klm	7.56	defghijklm
	Rusticano	96.67	abcd	7.26	defghijklmnopqr
	Scorsonera	86.67	ijkl	7.81	cdefghijk
	Cappelli	97.33	abc	8.01	cdefghi
	Simeto	95.33	abcdef	6.37	klmnopqrstuvwxy
	Timila reste nere	95.33	abcdef	9.53	ab
	Timilia reste bianche	91.67	abcdefghij	10.06	a
	Trinakria	82.00	lmn	6.69	hijklmnopqrstuvw
	Urria	93.33	abcdefghi	7.55	defghijklmn
50 mM	Capeiti 8	96.67	abcd	6.92	efghijklmnopqrstu
	Castiglione glabro	88.00	ghijkl	6.69	hijklmnopqrstuvw
	Ciciredda	88.67	fghijkl	6.99	defghijklmnopqrst
	Ciclope	95.33	abcdef	7.38	defghijklmnopq
	Duilio	92.00	abcdefghij	5.98	pqrstuvwxyz
	Francesa	91.33	bcdefghij	7.74	defghijkl
	K-26	92.67	abcdefghi	8.17	cdefg
	Mongibello	95.33	abcdef	6.55	ijklmnopqrstuvwx
	Perciasacchi	89.00	efghijk	6.04	opqrstuvwxyz
	Quadrato	92.00	abcdefghij	6.49	jklmnopqrstuvwx
	Russello	85.33	jkl	7.31	defghijklmnopq
	Russello Ibleo	92.67	abcdefghi	6.48	jklmnopqrstuvwx
	Rusticano	98.33	ab	7.42	defghijklmnop
	Scorsonera	87.33	hijkl	7.47	defghijklmno
	Cappelli	98.00	ab	8.40	bcde
	Simeto	94.00	abcdefgh	6.99	defghijklmnopqrst
	Timila reste nere	94.67	abcdefg	8.28	bcdef
	Timilia reste bianche	94.67	abcdefg	8.22	bcdefg
	Trinakria	74.00	o	6.55	ijklmnopqrstuvwx
	Urria	95.33	abcdef	6.99	defghijklmnopqrst
100 mM	Capeiti 8	94.00	abcdefgh	5.83	rstuvwxyz
	Castiglione glabro	88.67	fghijkl	6.18	mnopqrstuvwxy
	Ciciredda	92.67	abcdefghi	6.75	ghijklmnopqrstuvw
	Ciclope	98.00	ab	5.32	wxyz
	Duilio	90.67	cdefghij	5.14	xyz
	Francesa	90.00	defghijk	6.07	nopqrstuvwxyz
	K-26	95.00	abcdefg	6.00	opqrstuvwxyz
	Mongibello	96.67	abcd	5.77	stuvwxyz
	Perciasacchi	85.33	jkl	4.68	z
	Quadrato	87.33	hijkl	5.78	rstuvwxyz
	Russello	78.00	mno	5.56	tuvwxyz
	Russello Ibleo	78.33	mno	4.93	yz
	Rusticano	95.33	abcdef	6.27	lmnopqrstuvwxy
	Scorsonera	77.33	mno	6.02	opqrstuvwxyz
	Cappelli	95.00	abcdefg	6.54	ijklmnopqrstuvwx
	Simeto	94.67	abcdefg	5.50	uvwxyz
	Timila reste nere	98.67	a	6.84	fghijklmnopqrstuv
	Timilia reste bianche	96.00	abcde	7.88	cdefghij
	Trinakria	76.00	no	5.90	qrstuvwxyz
	Urria	94.00	abcdefgh	5.44	vwxyz

In each column, means followed by the same letter are not statistically different according to Duncan's Multiple Range test at the 5% probability level.

4. Discussion

Seed germination is a major factor limiting the establishment of plants under saline conditions. As supported by the available literature [4–7], our results confirm that durum

wheat seeds tend to germinate at a lower rate and take a longer time when exposed to salt stress. Increasing NaCl concentrations led to a rise in mean germination time and shoot dry matter and a reduction in the shoot length of the seedlings.

The interaction of wheat genotypes with salinity level was found to be significant for germination and root length. This means that there was a crossover effect among the cultivars tested for these parameters.

Among the genotypes, all the landraces showed the best performance in shoot length, and many of them have been signaled for good performance in germination and root length (Timilia reste bianche, Timilia reste nere, Ciciredda and Cappelli).

The root number value being significantly higher at 50 mM is probably due to a stimulating action of the solution, to be investigated in further studies. On the other hand, the increase in the maximum concentration resulted in a statistically significant reduction in this parameter.

In conclusion, we can affirm that landraces are interesting genetic materials to be investigated and used in breeding programs for the selection of varieties better tolerant to salt stress. Timilia reste bianche, Timilia reste nere, Ciciredda and Cappelli are highlighted for the best root length performance at 100 mM.

To confirm our results and extend the study at later growing stages, further study is needed.

Supplementary Materials: The presentation materials can be downloaded at: https://www.mdpi.com/article/10.3390/IOCAG2023-16339/s1.

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