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Evaluation of salinity tolerance indices in North African barley accessions at reproductive stage

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Abstract: Soil salinity is one of the main factors limiting cereal productivity in worldwide agriculture. Exploitation of natural variation in local barley germplasm is an effective approach to overcome yield losses. Three gene pools of North African *Hordeum vulgare* L. grown in Tunisia, Algeria and Egypt were evaluated at the reproductive stage under control and saline conditions. Assessment of stress tolerance was monitored using morphological, yield-related traits and phenological parameters of reproductive organs showing significant genetic variation. High heritability and positive relationships were found suggesting that some traits associated with salt tolerance could be used as selection criteria. The phenotypic correlations revealed that vegetative traits including shoot biomass, tiller number and leaf number along with yield-related traits such as spike number, one spike dry weight, grain number/plant and grain number/spike were highly positively correlated with grain yield under saline conditions. Hence, these traits can be used as reliable selection criteria to improve barley grain yield. Keeping a higher shoot biomass and longer heading and maturity periods as well as privileged filling ability might contribute to higher grain production in barley and thus could be potential target traits in barley crop breeding toward improvement of salinity tolerance. Multiple selection indices revealed that salt tolerance trait index provided a better discrimination of barley landraces allowing selection of highly salt-tolerant and highly productive genotypes under severe salinity level. Effective evaluation of salt tolerance requires an integration of selection indices to successfully identify and characterize salt tolerant lines required for valuable exploitation in the management of salt-affected areas.

Keywords: grain yield; *Hordeum vulgare*; morphological traits; phenological parameters; salt tolerance; selection indices

Salinity is gaining major importance worldwide due to climate changes and inappropriate management of irrigation practices. This abiotic limitation was extremely intensified with water deficit in arid and semi-arid areas over the world and especially within North Africa and Mediterranean Basin. Exploring the natural diversity of major economically cereal crops in these regions to select autochthonous salt-tolerant genotypes was considered as a promising strategy for crop improvement (Shahbaz & Ashraf 2013). Although barley (*Hordeum vulgare* L.) is moderately salt-tolerant, a wide genetic variation has been reported (Munns *et al.* 2006) and is highly required to survey barley germplasm. Investigating the response of several accessions in salt-affected environment will help to enhance the comprehension

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of key mechanisms for stress tolerance. Improving salinity tolerance involves a screening-based method to better exploit appropriate stress tolerance traits (El-Hendawy *et al.* 2009). Several selection indices have been suggested on the basis of a mathematical relationship between control and stress conditions. Despite of current efforts intended for assessing tolerance criteria based on tolerance indices in cereal crops during germination and seedling stage (Askari *et al.* 2016), little has been reported at maturity (Sardouie-Nasab *et al.* 2014). Furthermore, this tolerance undoubtedly appears to be stage-dependent (Akram *et al.* 2002) and must be evaluated at the yielding phase.

Salinity stress might affect spikelet initiation that can seriously influence quality and grain yield through modeling heading date. In recent years, heading in small grain cereals is an important stage that has been extensively studied and considered as highly related to environmental adaptation (Ibrahim *et al.* 2016; Alqudah & Schnurbusch 2017). For these reasons, ongoing breeding programs target different tolerance traits at phenological, morphological and agronomic levels.

This is the first report integrating heritability, relationship linking vegetative and yield-related traits as well as phenological characters, within multiple tolerance indices and investigating the patterns of genetic diversity in order to conserve and exploit efficiently North African barley germplasm. Since the study covered mainly landraces adapted to harsh environment (Allel *et al.* 2016), the challenge is of substantial concern in efforts to determine key morphological and agronomic traits associated with salt tolerance.

MATERIAL AND METHODS

Plant material, experimental design and salt treatment. In order to determine high yielding-related traits of North African barley, a total of 31 barley accessions and varieties collected from Tunisia (11), Algeria (9) and Egypt (11) were evaluated. A pot experiment was conducted from December to June at the Centre of Biotechnology in Borj Cedria (30 km, south of Tunis) under open experimental station equipped with a shelter to avoid rainfall interference. Pots were arranged in completely randomized design with two factors (salinity level and accessions) and nine replications, which gives a total of 837 pots (31 accessions × 3 treatments × 9 replicates). The seedlings were grown in 5l-pots filled with sandy loam soil and irrigated on a regular basis twice a week with half-strength Hoagland's nutritive solution. At 4-leaf developmental stage, salt treatment was initiated gradually until the final NaCl concentration was reached; 100 or 200 mM NaCl for moderate and high salinity levels respectively, while the control plants were irrigated with distilled water. The plants were harvested at maturity 15 weeks after starting salt treatment.

Data collection and estimation of salt tolerance indices. The accessions were assessed by means of phenological and morphological characterization at reproductive stage in all barley genotypes. Morphological data were recorded for 28 traits listed with their abbreviations in Table 1 including growth and reproductive characters. Plant reproductive phenology was monitored by determining the days to heading (HEA) and days to maturity (MAT). HEA were calculated as the number of days between the sowing date and the date when 50% of all the shoots in a plot had fully emerged spikes. MAT were calculated from sowing date to 50% senescence of the spikes.

Salinity susceptibility and tolerance indices were calculated based on grain yield production as follows:

Stress Tolerance Index (STI):

$STI = YP \times YS/(YAP)^2$ (Fernandez 1992)

Stress Susceptibility Index (SSI):

$SSI = 1 - YS/YP/SI$ (Fischer & Maurer 1978)

Tolerance Index (TOL):

$TOL = YP - YS$ (Rosielle & Hamblin 1981)

Mean Productivity (MP):

$MP = YP + YS/2$ (Rosielle & Hamblin 1981)

Geometric Mean Productivity (GMP):

$GMP = (YS \times YP)^{½}$ (Fernandez 1992)

Harmonic Means (HARM):

$HARM = 2(YP - YS)/YP + YS$ (Baheri *et al.* 2003)

Salt Tolerance Trait (STTI):

$STTI = (YS/YP) \times 100\%$ (Chen *et al.* 2007)

Stress Intensity (SI):

$SI = [1 - (YAS/YAP)]$

where:

YP – seed yield from non-stressed pot of a given genotype
YS – seed yield from stressed pot of that genotype
YAS – average seed yield of all genotypes from stressed pots
YAP – average seed yield of all genotypes from non-stress pots.

Statistical analysis. Analysis of the effects of genotype, treatment and treatment × genotype interactions on measured traits was performed using Proc GLM in SPSS (Ver. 16, 2007). Duncan's multiple range tests were applied for all phenotypic mean value at a significant level of 0.05.

Table 1. Studied traits in barley genotypes including phenological, growth, grain yield traits and morphological traits ratio (total of 28 traits)

Trait		Abbreviation
Phenological traits		
1	days to heading	HEA
2	days to maturity	MAT
Growth traits		
3	whole dry weight/plant	WDW
4	shoot dry weight/plant	ShDW
5	root dry weight/plant	RDW
6	plant height	PH
7	tiller No./plant	TN
8	leaf No./plant	LN
Grain yield traits		
9	grain yield/plant	GY
10	spike dry weight/plant	SDW
11	spike dry weight	1SDW
12	spike No./plant	SN
13	grain No./plant	GN/P
14	spikelet No./spike	StN
15	grain No./spike	GN/S
16	awn length	AL
17	rachis length	RL
18	spike length	SL
19	1000-kernel weight	TKW
Morphological traits ratio		
20	shoot/root dry weight	Sh/R
21	root/shoot dry weight	R/Sh
22	leaf/tiller No.	LN/TN
23	spike/tiller No.	SN/TN
24	spike/shoot dry weight	S/Sh
25	grain yield/shoot dry weight	GY/Sh
26	grain yield/whole plant dry weight	GY/W
27	grain yield/shoot and root dry weight	GY/Sh+R
28	harvest index	HI

The mean data of each trait for a given genotype and salt treatment level were used to calculate genotypic co-variance (σ^2_g), environmental co-variance (σ^2_g) and broad-sense heritability (H^2_b). H^2_b was calculated for each trait as

$$H^2_b = \sigma^2_g/(\sigma^2_g + \sigma^2_e)$$

Phenotypic correlations between growth and agronomic traits as well as phenological characters were estimated by computing the Person's correlation coefficient (*r*) using the SPSS Correlate procedure.

RESULTS AND DISCUSSION

Variance analysis. The North African barley germplasm was recently shown to provide a great potential for crop improvement (Allel *et al.* 2017). Systematic approaches to increase the level of stress tolerance require the evaluation of the existing genetic variability in barley gene pool. ANOVA showed highly significant difference between the genotypes, the treatment and their interaction in terms of agro-morphological and phenological characters that can explain the phenotypic trait variation (Table 2). Similar results were reported in bread wheat under imposed water deficit (Habibpour *et al.* 2012).

Generally, the greatest effect was observed for treatment (Table 2) as found in wheat (Borrelli *et al.* 2011). Regarding phenological traits, the genotype marked the major effect when compared to the treatment (Table 2).

Assessment of trait heritability. In this study, among all attributes, very high values of heritability (80–95%) were shown with HEA, MAT followed by GN/S, GY/ShDW, HI over the salt and the control treatments as well as AL, RL, SL under control condition (Table 3). Moderately high heritability was detected for WDW, PH, SN/S, Sh/R, R/Sh, GY/W, GY/Sh+R under saline and non saline condition; length of different part of the spike traits under saline condition as well as SDW and S/Sh under control condition. Heritability for the other traits were moderate like GY except for some traits that show low heritability (≤ 40%) including TN, SN, TKW, SN/TN under saline and non-saline condition as well as LN under 200 mM NaCl. According to Ahmadi *et al.* (2016), high estimates of heritability

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Table 2. Variance analysis (mean square and *F* value) of phenological characters, growth parameters, grain yield traits and morphological ratios in response to salinity stress

Phenological and growth traits

	df	days to heading	days to maturity	whole dry weight/plant	shoot dry weight/plant	root dry weight/plant	plant height	tiller No./plant	leaf No./plant
Mean square									
G	30	1839.03***	1931.16***	148.64***	31.22***	1.33***	679.10***	11.83***	1021.73***
T	2	311.63***	126.80***	1354.97***	167.19***	14.89***	3539.99***	94.15***	7673.05***
G × T	60	331.56***	325.29***	24.37***	4.35***	0.26***	87.88***	3.09***	150.29***
Error	744	0.20	0.21	3.01	0.99	0.05	18.62	0.94	55.31
F									
G		8981.91	8961.22	49.34	31.43	25.27	36.47	12.65	18.47
T		1522.00	588.41	449.8	168.3	282.29	190.11	100.62	138.73
G × T		1619.37	1509.44	8.09	4.38	4.87	4.72	3.3	2.72

Grain yield traits

	df	grain yield/plant	spike dry weight/plant	spike dry weight	spike No./plant	grain No./plant	spikelet No./spike	grain No./spike	awn length	rachis length	spike length	1000-kernel weight
Mean square												
G	30	21.71***	34.04***	3.35***	4.99***	16811.20***	37.05***	1333.76***	50.77***	30.02***	87.78***	434.54***
T	2	258.28***	347.26***	6.63***	59.75***	85497.25***	16.96**	610.60**	69.59***	15.90***	69.07***	4627.86***
G × T	60	5.99***	8.64***	0.43***	2.65***	2485.49***	12.13***	436.60***	5.32***	2.10***	8.07***	106.86***
Error	744	0.91	1.56	0.104	0.71	221.23	3.8	136.74	0.53	0.3	0.81	11.22
F												
G		23.82	21.82	32.16	7.01	75.99	9.75	9.75	96.61	99.24	107.83	38.72
T		283.47	222.63	63.6	83.84	386.47	4.47	4.47	132.44	52.54	84.84	412.37
G × T		6.57	5.54	4.16	3.72	11.24	3.19	3.19	10.13	6.95	9.91	9.52

Morphological traits ratio

	df	shoot/root dry weight	root/shoot dry weight	leaf/tiller No.	spike/tiller No.	spike/shoot dry weight	grain yield/shoot dry weight	grain yield/whole plant dry weight	grain yield/shoot & root dry weight	harvest index
Mean square										
G	30	11.17***	0.02***	29.02***	0.19***	2.15***	2.15***	0.12***	1.46***	0.12***
T	2	119.58***	0.24***	22.89***	0.05 ns	0.67***	1.41***	0.11***	1.25***	0.12**
G × T	60	13.11***	0.02***	9.62***	0.06***	0.30***	0.30***	0.03***	0.19***	0.02***
Error	744	0.63	0.001	1.87	0.02	0.074	0.019	0.003	0.015	0.003
F										
G		17.75	24.59	15.52	9.4	29.25	110.82	36.69	94.5	38.35
T		189.93	237.28	12.24	2.42	9.13	72.46	33.49	81.14	40.44
G × T		20.83	21.06	5.14	2.72	4.08	15.56	8.44	12.15	8.09

Significance levels: *$P \leq 0.05$; **$P \leq 0.01$; ***$P \leq 0.001$; ns – no significant; *F* – coefficient of Snedecor-Fisher; df – degrees of freedom; G – genotype; T – treatment were shown only for the GN/S (96%), TKW (75%), peduncle length (68%) and HEA (72%), while low heritability as shown for GY (24%).

In this study, high estimates of heritability were shown for several yield components, while GY had moderate heritability suggesting that the treatment effect constitutes a major portion of the total phenotype variation in this character. Thus, the selection of superior genotypes based on this character would not be effective. For a more efficient approach towards improvement of GY, selection should be made on its components. Moderate to low values of heritability for GY in barely and other crops have been reported previously (Chand *et al.* 2008; Ahmadi *et al.* 2016).

Additionally to the high heritability found here for several determined traits, data showed that the ratio between the coefficients of genetic and environmental variation was often above one for many characters over all treatments (Table 3). Both of those results suggest a high genetic control of the expression of these traits. Thus the selection can safely be performed on the basis of phenotypic expression of those characteristics in the individual plant by adopting simple selection methods.

Values of heritability were higher under control condition than under high salinity for the majority of traits (60% of the total studied traits); this finding is due to a higher genetic variance for these traits under control conditions and to a higher environmental variance for the remaining traits under salt treatment. In contrast, PH, GY/Sh, GY/W, HI showed higher heritability under high salt treatment compared to control condition.

Means and range of salt tolerance indices of agro-morphological traits. Based on agro-morphological evaluation, barley genotypes react differently to the salt treatment (Table S1 in Electronic Supplementary Material (ESM)). Mainly, results showed reduction of almost all vegetative traits (including ShDW and TN) as well as GY and its components (including SN, GN/P, GN/S and TKW) with increasing salinity level as compared to control plants. The salt-induced decline in growth and GY has been reported previously in rice (Mahmood *et al.* 2009), wheat (Borrelli *et al.* 2011) and barley (Ahmed *et al.* 2013).

The range and mean values of salt tolerance indices for all the studied characters also indicated wide ranges of variation which revealed possible amount of variability among the genotypes (Table S1 in ESM). For instance, high range of variability was observed at 100 mM NaCl for SDW followed by RDW. In addition, wide range of variability was shown at 200 mM NaCl for StN and GN/S and GY. Similarly, considerable variation of salt tolerance indices for agro- morphological traits was detected in rice under saline condition (Senguttuvel *et al.* 2016).

Salt-induced changes in pheno-phases duration. Phenological data for HEA and MAT revealed a notable difference between cultivars at control and 200 mM NaCl treatments (Table 4). The variability in maturation made it possible to identify early landraces as Early1 (135 days) and late landraces like Tichedrett and Temassine (181 days) at control condition. Salt-induced changes in pheno-phases duration have been reported in cereal crops (Hamam & Negrim 2014) and could have a huge impact on crop yield (Gol *et al.* 2017).

Salinity which tends to shorten the grain filling period and accelerates maturation will significantly reduce the final grain weight (Maas & Grieve 1990). Interestingly we found that at severe salinity (200 mM NaCl), HEA and MAT exhibited an increase in the majority of moderately and salt tolerant genotypes (based on STTI of GY; STTI ≥ 50%) suggesting that in these cultivars a longer HEA and MAT periods may contribute to salt tolerance. Thus, we deduce that delayed heading and maturity processes gives the opportunity of late differentiation and ripening, allowing the plant to maintain higher number of kernels/spike and consequently high GY.

Relationships between growth, yield and its related components and phenological parameters. Since the present study aims to provide clues to identify the most desirable performance criteria as alternatives for breeders to increase yield, relationships between agro-morphological (Table S2 in ESM) and phenological traits (Table S3 in ESM) were monitored in barley. GY is often proposed as the primary trait for this selection designed for achieving adaptation to stress-prone environments (Araus *et al.* 2008). Interestingly we found that ShDW, WDW, LN, TN were strongly and significantly correlated with GY. Thus, we recommend these vegetative components as suitable parameters to assess for high GY and salt tolerance as they permit direct estimation of economic return under saline conditions. Similarly, some studies proposed TN and ShDW as valuable criteria to evaluate salt tolerance (Ahmed *et al.* 2013; Sbei *et al.* 2014). It was reported that in cereals salt can inhibit tiller and leaf formation during their emergence as well as it may even accelerates leaf senescence and abortion of tiller which results in reducing green biomass and GY (Grieve *et al.* 2001).

Furthermore, our results indicated that under saline condition the decline of GY was positively correlated with ShDW decline. A decrease in photo-

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Table 3. Heritability values (mean square) of growth parameters, grain yield traits and morphological ratios under control condition (without NaCl) and in response to high salinity stress (200 mM NaCl)

Phenological and growth traits

	days to heading	days to maturity	whole dry weight/plant	shoot dry weight/plant	root dry weight/plant	plant height	tiller No./plant	leaf No./plant
Control								
σ^2_g	103.41	107.32	10.01	1.99	0.09	29.98	0.58	49.37
σ^2_e	4.87	10.66	3.62	1.54	0.08	20.27	1.05	60.43
H^2_b (%)	95.31	91.25	73.45	56.45	53.76	59.65	35.53	44.96
200 mM NaCl								
σ^2_g	122.63	126.20	3.947	0.84	0.03	28.99	0.37	31.82
σ^2_e	9.80	14.09	2.49	0.71	0.03	16.95	0.76	56.59
H^2_b (%)	93.33	90.12	61.32	54.3	52.31	63.1	32.74	35.99

Grain yield traits

	grain yield/plant	spike dry weight/plant	spike dry weight	spike No./plant	grain No./plant	spikelet No./spike	grain No./spike	awn length	rachis length	spike length	1000-kernel weight
Control											
σ^2_g	1.661	2.415	0.187	0.388	600.32	1.71	64.705	2.546	1.277	4.38	12.01
σ^2_e	1.34	1.93	0.1	0.73	615.41	0.54	10.51	0.52	0.32	0.98	26.34
H^2_b (%)	55.35	55.58	64.71	34.8	49.38	76	86.03	83.09	79.81	81.76	31.32
200 mM NaCl											
σ^2_g	0.63	0.89	0.11	0.15	597.72	1.11	47.03	1.82	1.07	2.43	15.23
σ^2_e	0.6	1.19	0.099	0.54	601.92	0.37	8.39	0.63	0.28	0.83	31.24
H^2_b (%)	51.02	42.87	51.47	22.19	49.83	75.12	84.86	74.25	78.98	74.44	32.77

Morphological traits ratio

	shoot/root dry weight	root/shoot dry weight	leaf/tiller No.	spike/tiller No.	spike/shoot dry weight	grain yield/shoot dry weight	grain yield/whole plant dry weight	grain yield/shoot and root dry weight	harvest index
Control									
σ^2_g	1.264	0.003	1.991	0.012	0.113	0.096	0.006	0.062	0.095
σ^2_e	0.43	0.001	1.87	0.02	0.06	0.022	0.004	0.02	0.021
H^2_b (%)	74.57	75	51.63	40	63.84	81.36	60	79.49	80.33
200 mM NaCl									
σ^2_g	1.42	0.003	1.73	0.01	0.07	0.093	0.01	0.06	0.092
σ^2_e	0.69	0.001	1.92	0.021	0.092	0.017	0.003	0.015	0.016
H^2_b (%)	67.49	75	47.44	25	41.77	84.55	66.67	78.57	85.19

h^2_b – heritability in broad sense; σ^2_g – genotypic variance; σ^2_e – environmental variance

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synthetic rate during vegetative (Allel *et al.* 2018) and reproductive phase (Hirasawa *et al.* 2017) as well as leaf injury caused by the salt toxicity could explain a reduction in biomass that affects later GY production.

In order to give a clear estimation of GY production versus total biomass or above-ground biomass, HI and GY/vegetative traits ratio were determined. Data showed that these agronomic traits were positively associated with GY under salinity indicating that plants able to maintain HI under stress conditions will often have a higher yield.

Here, our results revealed that growth vigor is one of the major determinants for high yield performance in barley. Accordingly, Kumar *et al.* (2013) and Reddy *et al.* (2017) suggested that plant vigor helps to avoid the toxic effects of salinity rather than stands as a tolerance mechanism which works as far as the productivity is concerned.

In the present study, we found that yield components including SN, 1SDW, GN/P, GN/S, Grain Number/Rank (GN/R) and TKW were positively correlated with GY under saline condition. Thus we assumed that these characters are best predictors of yield under high stress conditions. This finding is supported by authors (Ji *et al.* 2012; Akbarpour *et al.* 2015) who noted similar behavior in wheat exposed to salt stress and in rice subjected to drought condition, respectively. In addition, we noticed that correlation coefficient of the grain filling parameters cited above and TKW increased with increasing salinity level (Table S2 in ESM) suggesting a main role of the grain filling process in efficient GY under salt stress as well as a photosynthetic activity contribution could be anticipated. Data obtained on phenological and agro-morphological relations showed that GY and dry weight production of growth parts were significantly and positively correlated with HEA and MAT under saline and non-saline conditions (Table S3 in ESM).

In summary, the high yield is primarily associated with increased biomass and shoot growth which subsequentially allows better salinity tolerance in barley.

Estimation of tolerance indices. According to our data, GY was significantly reduced with increasing salinity except for some specific salt tolerant genotypes (Table S4 in ESM). The Giza125, Giza126, Temassine and Kebilli2 genotypes yielded the greatest dry weights at moderate salinity while, the lowest value was observed in the Early1 and Tombari genotypes. Many interesting genotypes were identified at high salinity, Tozeur2, Tichedrett, Kerkena and Kebilli2 were among the most productive, while El Arich, Tombari and Early1 genotypes were the less productive. For better evaluation of barley landraces for salt tolerance, seven selection indices including STI, MP, GMP, TOL, SSI, HARM and STTI were used (Table S4 in ESM).

In the present study, the maximum values of STI, MP and GMP were observed in Kebilli2, Temassine

Table 4. Effect of salinity on pheno-phases duration in barley phenological characters including number of days to heading (HEA) and number of days to maturity (MAT) were determined in barley under control and saline conditions (200 mM NaCl) (in days)

	Control		200 mM NaCl	
Genotypes	HEA	MAT	HEA	MAT
Kebilli 2	131^{j}	171^{h}	139^{d}	178^{d}
Tozeur 2	132^{i}	172^{g}	139^{d}	178^{d}
Rihane	117^{q}	155^{s}	117^{q}	155^{s}
Manel	131^{j}	168^{k}	131^{f}	168^{i}
Jerba	123^{m}	165^{m}	121^{o}	162^{n}
Sidi Bouzid	137^{d}	172^{g}	147^{b}	182^{c}
Kairouan	138^{c}	178^{d}	135^{e}	174^{f}
Gabès	139^{b}	178^{d}	130^{g}	169^{h}
Tozeur 1	129^{k}	169^{j}	121^{o}	160^{p}
Tombari	136^{e}	171^{h}	131^{f}	166^{k}
Kerkna	136^{e}	176^{e}	127^{j}	166^{k}
Sidi Mehdi	139^{b}	179^{c}	148^{a}	187^{a}
Ras El Mouch	121^{n}	161^{n}	148^{a}	187^{a}
Ksar Megarine	135^{f}	173^{f}	139^{d}	177^{e}
Saïda	139^{b}	176^{e}	129^{h}	166^{k}
Azrir	108^{r}	146^{u}	108^{r}	146^{t}
Rihane 03	133^{h}	173^{f}	135^{e}	174^{f}
Tichedrette	141^{a}	180^{b}	128^{i}	167^{j}
Nailia	119^{o}	158^{q}	119^{p}	158^{r}
Temassine	141^{a}	181^{a}	126^{k}	165^{l}
Early 1	100^{s}	135^{v}	100^{s}	135^{u}
Early 2	115^{q}	153^{t}	123^{m}	161^{o}
Giza 123	117^{p}	157^{r}	122^{n}	160^{p}
Giza 125	117^{p}	155s	121^{o}	149^{q}
Giza 126	127^{l}	169^{j}	130^{g}	171^{g}
Giza 127	119^{o}	159^{p}	124^{l}	163^{m}
Giza 129	132^{i}	172^{g}	146^{c}	185^{b}
Giza 130	134^{g}	170^{i}	146^{c}	182^{c}
Giza 131	127^{l}	167^{l}	127^{j}	166^{k}
Giza 2000	119^{o}	160^{o}	121^{o}	161^{o}
El Arich	121^{n}	159^{p}	121^{o}	159^{q}

Letters indicate significant differences at $P < 0.05$ according to Fisher’s test

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and Giza126 genotypes at 100 mM NaCl, Keblli2 and Tozeur2 at 200 mM NaCl indicating that these genotypes had high capacity of GY production at both stress and non stress conditions simultaneously. STI, MP and GMP take into consideration GY production of the genotype under both of control and salt condition allowing to identify the genotypes that perform well under both of these treatments. However, these cited indices may let to ineffective selection since GY might fluctuate whether plants were exposed to saline or non saline condition. For instance, the genotype Kairouan showing higher value of STI, MP and GMP appeared moderately salt tolerant genotype with an STTI value of 50.6% and not the most salt tolerant.

In the other hand, SSI, TOL, HARM and STTI indices take into consideration only the capacity of the genotype to sustain yield under salt stress compared to control condition and allowed to truly select the most salt tolerant genotypes.

Our results showed that the lowest values of SSI, TOL and HARM indicating the most salt tolerant genotypes were recorded for Giza125 and Giza130 at 100 mM and Saïda, Tichedrett, Early1, Azrir and Giza125 at high salinity level. Considering the STTI indices, the highest values indicated the most salt tolerant genotypes which were the same identified by SSI, TOL and HARM. Such finding suggests that these genotypes possess sufficient plasticity to respond to soil salinity constraint as well as an implication of significant salt tolerance mechanisms. Similar results were found by Senguttuvel *et al.* (2016).

If selection strategies deal with factors of stress adaptation in addition to yield under stress, then it may be possible to combine higher yield potential and salt resistance (Blum 2005).-Interestingly, using multiple tolerance indices, we have provided evidence for both high grain yield production and substantial tolerance at severe salinity in a quite number of genotypes like Tichedrett. In fact, from an evolutionary perspective throughout domestication and breeder selection over time, salt-responsive genes have made a contribution to the development of salt barley cultivars, especially in harsh environments (Allel *et al.* 2016, 2017).

The salt tolerance indices studied here provided different classifications and arranged the genotypes based on particular performance as illustrated in Figure S1 (in ESM) showing that STI, TOL, HARM and STTI appeared the most efficient selection indices with a clear variability based on GY, while SSI, MP and GMP failed to differentiate between genotypes. Several studies proposed the use of STTI in screening program for salt tolerance rather than other indices (Ali *et al.* 2007; Shahzad *et al.* 2012).

Approaches for sustaining high yields of barley under salinity are important agricultural goals. Various salt tolerance indices were shown to be useful for estimation of stress level and identification of salt tolerant elite genotypes. Such potential lines could serve for effective exploitation in breeding purposes or as source for salt-responsive candidate genes suitable for crop improvement in salt-affected soils.

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