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IMBIBITION, GERMINATION AND LIPID MOBILIZATION RESPONSE BY SUNFLOWER SUBJECTED TO SALT STRESS

ABDUL KABIR KHAN ACHAKZAI

Department of Botany, University of Balochistan, Quetta, Pakistan
Corresponding author e-mail: profakk@yahoo.com, Ph #: 081-9211264, Cell #. 0333-7812944

Abstract

Salinity is one of the most important abiotic stresses in arid and semi-arid regions that substantially reduce the germination, growth and average yield of major crops. The study was mainly aimed to select the most salt tolerant cultivar of sunflower. Therefore, a pot culture experiment was conducted to study the effects of four different salinity levels having electrical conductivity *viz.,* 1.19, 9.54, 16.48 and 22.38 mS/cm on the imbibition (water uptake), germination and lipid mobilization of seedlings of 4 varieties of sunflower (*Helianthus annuus* L.) i.e., DO-728, DO-730, Hysun-33 and Suncross-843. Salinity levels were prepared by dissolving calculated amount of NaCl, Na_2SO_4, $CaCl_2$ and $MgCl_2$ (4:10:5:1) in half strength Hoagland culture solution. Imbibition was studied using plastic glasses at an interval of 12 and 24 hours. While germination studies were separately carried out in plastic pots and noted after every 12 hours till 20 days. Whereas, lipid contents of the salt stress germinating seeds were determined at three time intervals *viz.,* 48, 96 and 144 hours of germination. Results showed that there was a linear decrease in imbibition, germination and lipid mobilization as the level of salinity progressively intensifies. Maximum significant reduction in imbibition (12.88%), germination (31.03%) and lipid mobilization (38.62%) is recorded in highest dose of applied salts (22.38 mS/cm). Results further exhibited that maximum significant reduction in imbibition (17.95%) and germination (43.05%) is recorded for variety Suncross-843. While minimum for the same attributes is recorded for variety DO-728. Therefore, in term of imbibitions and germination, DO-728 could be ranked as salt tolerant**.** Similarly maximum reduction (14.85%) in mobilized lipids is noted for DO-728 and minimum (40.89%) for DO-730. Therefore, in term of lipid mobilization, variety DO-730 could be ranked as salt tolerant and DO-728 as salt sensitive. While remaining 2 varieties i.e., Hysun-33 and Suncross-843 is rated as salt intermediate in response, respectively.

Key words: Sunflower, Salinity, Imbibition, Germination, Lipid mobilization, Seedlings.

Introduction

Salinity is one of the major abiotic stresses in arid and semi-arid regions that substantially reduce the average yield of major crops by more than 50% (Bray *et al.,* 2000; Ashraf, 2009). Salinity in soil or irrigation water is the major hindering factor for crop growth in many regions of the globe (Kausar *et al.,* 2013; Pirasteh-Anosheh *et al.,* 2014). Salt stress at any stage of crop growth can cause an irreversible loss in yield potential in many studied crop plants (Hameed *et al.,* 2013). However, seed germination and seedling establishment are the periods when crop is most sensitive to salt stress (Yagmur *et al.,* 2007; Emam, 2011). Like many other abiotic stresses, salt stress considerably reduces the growth and development of a number of plants by affecting their various physiological and biochemical processes (Shahbaz *et al.,* 2011, 2012; Habib *et al.,* 2012; Shahbaz & Ashraf, 2013). Crops growing in salt affected soils may suffer from physiological drought stress, ion toxicity, and mineral deficiency which lead to reduce germination, growth, and productivity (Valiollah, 2013; Aziz *et al.,* 2013). Excess salt may affect plant growth either through osmotic inhibition of water uptake by roots or specific ion effects, which may cause toxicity (Saqib *et al.,* 2012; Abbas & Akladious, 2013). The first stage in plant development is seed germination, which starts by water uptake (imbibition), followed by reserve mobilization and protein synthesis and ends with the emergence of radicle from seed tissues (Bewley, 1997). Research studies also indicated that most of the biochemical and molecular changes are intensified during the first hours of imbibition (Harb, 2012). Furthermore, germination and seedling stage is predictive of plant growth responses to salinity (Cuartero *et al.,* 2006). There was a regular decrease in seed germination and seedling growth raised in Petri dishes for ten days with increasing salt concentration (Jamil *et al.,* 2012). Therefore, seeds with more rapid germination under salt stress and/or normal conditions may be expected to achieve a rapid seedling establishment and more salt tolerance, resulting in good stand establishment and higher yield productivity (Munns, 2002). Seeds are particularly vulnerable to environmental stress encountered between sowing and seedling establishment (Carter & Chesson, 1996). The over-changing goal of crop establishment is to obtain quick and uniform germination, followed by rapid and uniform seedling emergence plus autotrophy. Poor germination and seedling establishment are the results of soil salinity (Covell *et al.,* 1986).

Seeds play a vital role as dispersal units, as well as sources of food reserve to sustain the development and establishment of seedlings, maintaining the diversity of plant species (Borghetti & Ferreira, 2000). Proteins, lipids and carbohydrates are the main components stored during the late stages of seed development. The major seed storage reserves in oilseeds are accumulated in protein and oil bodies, and serves as an energy, carbon and nitrogen source during germination and post germination events (De-Carvalho *et al.,* 2001; Zienkiewicz *et al.,* 2013). Therefore, information concerning how an embryo mobilizes its internal reserves during the early stages of germination can provide insights into the metabolic process of germination and consequently into the ability to use such seeds as planting material (Gonçalves *et al.,* 2003). There are very few reports on the effects of salinity on storage-lipid degradation of germinating oil seeds. Kayani *et al.,* (1990)

reported that salinity significantly reduced germination and the amount of lipid used in 'Vista' jojoba [*Simmondsia chinensis* (Link) C.K. Schneid.]. They also pointed out that higher levels of salinity resulted in delayed initiation of germination and lipid breakdown. There are many strategies to overcome the negative effects of salinity. A good one strategy is the selection of cultivars/varieties and species for salinity (Ashraf *et al.,* 1992). Difference among species and varieties/cultivars for salinity tolerance may reside in their differences in salinity tolerance mechanism. Exploitation of these useful genetic variations in salinity tolerance particularly of crop plants is an economical approach for proper utilization of salt-affected agricultural lands. In view of the above fact, the present study was therefore, conceived with to investigate the effects of varying level of salinity upon the imbibition (water uptake), germination and lipid mobilization of four different sunflower varieties/cultivars.

Materials and Methods

Present study deals with the effect of four different saline regimes (i.e., S_1, S_2, S_3 and S_4) having an electrical conductivity values of 1.19; 9.54; 16.48 and 22.38 mS/cm respectively on the imbibition (water uptake), germination and lipid mobilization of sunflower (*Helianthus annuus* L.). The certified seeds of four varieties of sunflower *viz.,* DO-728, DO-730, Hysun-33 and Suncross-843 were obtained from Agricultural Research Institute (ARI), Quetta. The above treatments were prepared by dissolving calculated amount of NaCl, Na_2SO_4, $CaCl_2$ and $MgCl_2$ (having ratio 4 : 10 : 5 : 1) in half strength Hoagland culture solution as explained by Machlis & Torrey (1956), and is shown in Table 1. The pH and the electrical conductivity of the treated solutions were determined using AGB-400/UP pH/conductivity and temperature meter.

Table 1. Amount of salt added in one-liter solution of various treatments.

Salinity treatments (S) EC = mS/cm	Amount of salts, g L^{-1}				Solution strength (mM)	pH
	NaCl	$Na_2SO_4.H_2O$	$CaCl_2$	$MgCl_2$		
S_1 (1.19)	-	-	-	-	-	4.03
S_2 (9.54)	1.17	3.2	2.35	1.9	20	4.40
S_3 (16.48)	2.34	6.4	4.70	3.8	40	4.36
S_4 (22.38)	3.51	9.6	7.05	5.7	60	4.30

Imbibition: The imbibition (water uptake) of seeds was studied using plastic glasses of 6 cm in diameter and 10 cm in depth. Twelve glasses were selected and marked, then filled with water washed sand. Each glass had drainage hole on its bottom. In each glass 3 healthy and uniform size seeds were sown. Before sowing each seed was weighed (W_1) and numbered. In each glass 15 ml respective salinity solution was added and placed on laboratory table at room temperature. After 12 hours, these seeds were taken out from the sand and dried on filter paper and reweighed (W_2). The seeds were then once again sown at their own places for next 12 hours. So after 12 more hours these seeds were again taken out, dried on filter paper and reweighed. The imbibition percentage was then calculated with the help of formula given below:-

$$\text{Imbibition (\%)} = \frac{W_2 - W_1}{W_1} \times 100$$

Germination: The germination studies were carried out in plastic pots of 17.5 cm in diameter and 6.5 cm deep having drainage hole on its bottom. Twelve pots were used for each variety and marked. Then filled with water washed sand and labeled as S_1, S_2, S_3, and S_4. Each salinity treatment was replicated thrice for each variety of sunflowers. More or less uniform size and equal number of seeds were sown in each pot at equidistant and equal amount i.e., 50 ml respective saline solution was also added in each pot. These pots were then placed on laboratory tables at room temperature for 20 days. The observations regarding germination were made after every 24 hours. The emergence of radical/plumule was taken as an index of germination.

Lipid extraction: Moreau & Huang (1977) method for lipid extraction was used. A 5.0 g fresh non-germinated seeds of each variety of sunflowers were taken and their testa (seed coat) was removed separately. Seeds were then grinded with methanol. After 20 minutes 100 ml chloroform was added in it. This solution was then transferred into a beaker and left for two hours at room temperature. After 2 hours, it was filtered and its filtrate was then transferred into a separating funnel by adding 2:1 water into it. Two layers were formed; the lower layer was collected in a beaker. This beaker was kept in an oven for 24 hours at 80°C. After 24 hours the beakers of each variety was weighed and subtracted the initial weight of the beaker and then calculate the total lipid contents (control) of each variety. Thereafter, lipid contents of the germinating seeds subjected to various levels of salt stress were determined at three time intervals *viz.,* 48, 96 and 144 hours of germination. The lipid extraction procedure for germinating seedlings was the same as adopted for non-germinating (control) seeds.

Statistical analyses: Data obtained were statistically analyzed for analysis of variance (ANOVA) and multiple comparison tables for various traits (i.e., imbibition, germination and lipid mobilization) using computer software Statistix version 8.1 (2005) to determine the effect of salinity (S), varieties (V) and time (T). Two factor interactions (S x V; S x T and V x T) and three factor interactions (V x S x T) was also determined. Their least significant differences (LSDs) were manually calculated with the help of formulae given in Table 2.

Table 2. LSDs formulae at probability level α for cell and marginal means in a three-factor factorial design for three different traits of sunflower subjected to four levels of salt stress.

Salinity (S) = a levels, Varieties (V) = b levels, Time (T) = c levels.							
	S	**V**	**T**	**S x V**	**S x T**	**V x T**	**S x V x T**
Traits $^{\Psi}$	$t^{**}_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE^{*}}{rbc}}$	$t_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE}{rac}}$	$t_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE}{rab}}$	$t_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE}{rc}}$	$t_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE}{rb}}$	$t_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE}{ra}}$	$t_{\frac{\alpha}{2}(\upsilon)}\sqrt{\frac{2\,MSE}{r}}$

* MSE = Pooled Mean Square Error, ** Critical t-value at α probability level

$^{\Psi}$ Traits (Imbibitions: a = 4, b = 4, c = 2, Germination: a = 4, b = 4, c = 2, Lipid Mobilization: a = 4, b = 4, c = 3)

Results and Discussion

Results showed that in response to various levels of salinity (S), imbibition, germination, and lipid mobilization of all varieties (V) with respect to different time intervals (T) are statistically exhibited highly significant results ($p<0.01$). Interaction between S x T; V x T; S x V and S x V x T were also found highly significant (Table 3). Similar results have also been obtained by Achakzai *et al.,* (2010a&b) for macro and micronutrients uptake by sunflower varieties of the present set of experiment.

Imbibition (%): Data showed that there was a significant linear decrease in imbibition (%) of sunflower subjected to different levels of salt stress. A maximum decline in imbibition (12.88%) is recorded in highest dose of salinity (22.38 mS/cm), while minimum (20.39%) is noted for salt free treatment (Table 4). The cultivars also showed significantly negative response in all salt regimes. A maximum reduction (14.57%) is recorded for Suncross-843 and minimum for DO-728. Results further exhibited that time had a great influence on the imbibition process of seed. A 24 h period significantly showed greater rate of imbibition over 12 h periods in case of all cultivars. The first step in the germination of any seed is imbibition, or water uptake, during which water needed for the biochemical and molecular changes in the germinating seed, is absorbed. Imbibition during the first 24 hours (h) for few seeds and 48 h for others is very crucial for the success of seed germination process (Harb, 2012). Any seeds absorb water primarily through the micropyl/or seed coat. During the first 12 h of the imbibition process, seeds may raise in water contents to between 30 and 40%. The hydration of large molecules within the seed causes water uptake; this occurs until the molecules are restored to full turgor. The pressure exerted against the cell wall by the increase water content causes the seed cells to become more turgid (rigid). A very little is known about salinity and seed imbibition. However, few researchers stated that rate of water uptake in rice seeds were reduced in increasing salinity, and rice grains attained full imbibition by 48 h up to 150 mM salinity. Salinity as an abiotic hazard induces several disorders in seeds and propagules during germination. It first reduces imbibition of water because of the lowered osmotic potential of the imbibing medium. Therefore, present findings of imbibition are in line with the results obtained by Alam *et al.,* (2003). The cultivars also showed significant response in relation to various levels of salt stress. A maximum reduction (14.57%) is recorded for Suncross-843 and minimum (17.95) for DO-728 (Table 4).

The results obtained for pair-wise comparisons test of each 2 factor interaction mean (i.e., S x V; S x T and V x T) also deciphered that under each pair of variables a maximum imbibition is recorded for non-stress treatment (22.91%), variety DO-728 (22.42%), and time 24 h (23.99%). While reverse is true as salinity level increases (Table 7abc).

Germination (%): Results pertaining to sunflower germination exhibited that there is a linear significant ($p<0.01$) decline after every rise in salinity level (Table 5). A maximum reduction (31.03%) is recorded in highest dose of applied salts (22.38 mS/cm). Results also exhibited that as the time period (DAS) increases the germination (%) also progressively increased within the same level of salt stress. However, such an increase is comparatively found lesser in treatments receiving higher concentration of salts. As we know germination is the process whereby the dormant seeds become active and begin the production of new cells. The over changing goal of crop establishment is to obtain quick and uniform germination followed by rapid and uniform seedlings. Poor germination and seedling establishment are the results of soil salinity. Researchers revealed that the rate of germination was significantly lower under saline conditions. They also noted that however, the germination percentage decreased and the time to germination also increased (delay in germination) with an increase in salt stress. Therefore, our present findings of germination are strongly in agreement with the results obtained and explained by these researchers (Singh *et al.,* 2004; Bahrani & Niknejad-Kazempour, 2007; Yağmur *et al.,* 2007; Necajeva & Levinish, 2008; Tlig *et al.,* 2008; Yağmur & Kaydan, 2008; Shereen *et al.,* 2011). Generally salt stress causes both osmotic and ionic stress. By decreasing the osmotic potential of the soil solution, seed/plant access to soil water is decreased, because of the decrease in total soil water potential. Therefore, higher osmotic pressure or saline conditions could be detrimental to the sunflower seeds.

Results also deciphered that under salt stress, sunflower cultivars responded significantly. Highest seed germination (54.58%) is recorded for cultivar DO-728 and a minimum (43.05%) for Suncross-843. While cultivar DO-730 and Hysun-33 are intermediate in germination response toward salt stress. This reduction is more substantial in higher than in lower concentration of applied salts. Similar trend of results for cultivar under salt stress are also obtained by many other researchers (Ashraf *et al.,* 1992; Okçu *et al.,* 2004). There are many strategies to overcome the negative effects of salinity. A good one strategy is the selection of cultivars/ species for salt tolerance. However, tolerance potential of different species varies greatly within species or even among different genotypes of the same species. Therefore, among four sunflower cultivars, DO-728 could be used as a salt tolerant cultivar.

Table 3. Analysis of variance (ANOVA) for imbibition (IMB), germination (GRN) and lipid mobilization (LPM) by four varieties of sunflower (*Helianthus annuus* L.) subjected to various levels of salinity.

Source	df			MS			F-Values		
	IMB	GRN	LPM	IMB	GRN	LPM	IMB	GRN	LPM
Salinity (S)	3	3	3	240.390	12728.6	1125.41	1216.84**	161385**	1005.31**
Varieties (V)	3	3	3	49.445	1332.9	1342.10	250.29**	16899.3**	1198.88**
Time (T)	1	3	2	957.228	31546.8	2098.37	4845.44**	399980**	1874.44**
S x T	3	9	6	2.670	658.3	37.42	13.51**	8346.62**	33.42**
V x T	3	9	6	19.481	80.9	38.16	98.61**	1025.24**	34.08**
S x V	9	9	9	2.964	124.6	44.62	15.01**	1580.31**	39.86**
S x V x T	9	27	18	1.591	39.70	7.89	9.16**	503.43**	7.05**
Error	64	128	96	0.198	0.10	1.12			
Total	**3**	**3**	**3**						

**Data is significant at p<0.01

Table 4. Effect of salinity on imbibition (%) of four varieties of sunflower (*Helianthus annuus* L.) at two different times.

Salinity treatments (S)	DO-728		DO-730		Hysun-33		Suncross-843		Salinity means
	12 Hours	24 Hours	12 Hours	24 Hours	12 Hours	24 Hours	12 Hours	24 Hours	
S_1	17.23 jk	28.50 a	17.553 j	23.890 b	16.587 kl	22.393 c	15.710 lm	21.193 d	20.394 a
S_2	14.740 no	23.437 b	15.593 mn	20.007 ef	14.607 o	19.207 fg	12.427 qr	18.500 ghi	17.315 b
S_3	12.147 qr	20.297 de	13.457 p	18.037 hij	13.393 p	18.687 gh	9.900 s	17.607 ij	15.440 c
S_4	9.703 s	17.450 jk	11.677 r	16.392 lm	10.587 s	16.050 lm	8.190 t	12.993 pq	12.880 d
Varietal means	**17.950 a**		**17.075 b**		**16.439 c**		**14.565 d**		**16.507**

LSD (p<0.01) values for salinity (S), varieties (V), time (T) and for interactions (S x V x T) are 0.341, 0.341, 0.241 and 0.965, respectively

Mean values followed by the same letter(s) within right side column (salinity) and bottom row (varieties) of the Table are not significantly different (p<0.01) using

LSD test. Similarly, values followed by the same letter(s) within column and rows (salinity treatments x varieties) in the center of the Table are not significantly different from each other

The results obtained for pair-wise comparisons test of each 2 factor interaction mean (*viz.,* S x V; S x T and V x T) also deciphered that under each pair of variables a maximum germination percentage is recorded for non-stress treatment (72.49%), variety DO-728 (78.73%), and time 20 DAS (100%). While reverse is found as salinity level increases (Table 7abc).

Lipid mobilization (%): Results regarding lipid mobilization deciphered that as the concentration of salt progressively increases the lipid mobilization (%) of soaked sunflower seeds inversely decreases (Table 6). A maximum amount of immobilized lipid contents (38.62%) is recorded in highest dose of applied salts (22.38 mS/cm). Results also deciphered that as the time period proceeds on, lipid mobilization also progressively increased within the same dose of applied salts, and a maximum response is noted at 144 h after sowing. As we know that lipid is the main source of food stored in the endosperm of sunflower seeds. After being seed imbibed, water allows chemical reaction to proceed in cells and to bring about the conversion of stored food into simple sugars and amino acids through enzymatic actions. These are then transported to the embryo, where some are respired to provide energy for germination and post-germination events and others are used in the synthesis of protein, starch and cellulose. Water and oxygen are necessary for the enzymes to break down the seed reserves. Greater the rate of mobilization of stored food, faster would be the rate of energy production for the newly developing cells providing that no environmental factor would hinder these chemical processes. However, studies revealed that salinity affects several aspects of plant metabolism, including lipid mobilization, and the effects of salinity have been studied at various stages of plant growth. Though much is known about the effect of salinity on crop plants, but very few works have dealt with the physiology of oilseeds. Hence, based on available literature, studies revealed that lipid metabolism exhibited a negative linear behavior, decreasing 40% between the first and last stage analyzed. Therefore, our present findings of lipid mobilization in response to salt stress are strongly in conformation with the results recorded by other investigators (De-Carvalho *et al.,* 2001; Gonçalves *et al.,* 2003; Lima *et al.,* 2008).

Table 5. Effect of salinity on germination (%) of four varieties of sunflower (*Helianthus annuus* L.) at four different interval times of days after sowing (DAS).

Salinity treatment (S)	DO-728				DO-730				Hysun-33				Suncross-843				Salinity means
	5 DAS	10 DAS	15 DAS	20 DAS	5 DAS	10 DAS	15 DAS	20 DAS	5 DAS	10 DAS	15 DAS	20 DAS	5 DAS	10 DAS	15 DAS	20 DAS	
S_1	30.00r	53.34l	100.00a	100.00a	30.00r	60.00j	100.00a	100.00a	25.00s	50.00m	96.77b	100.00a	25.02s	50.00m	93.33c	100.00a	69.590 a
S_2	30.00r	50.00m	70.00g	80.00e	23.33t	50.00m	66.66h	83.33d	20.00u	46.66n	60.00j	73.33f	20.00u	46.66n	60.22j	73.33f	53.345 b
S_3	16.66v	46.65n	63.33i	70.00g	20.00u	46.66n	56.66k	70.00g	16.00w	33.33q	50.00m	66.55h	13.33x	20.00u	46.55n	60.30j	43.504 c
S_4	10.00y	33.33q	56.66k	63.33i	0.00z	33.33q	43.33o	60.00j	0.00z	19.98u	40.00p	56.45k	0.00z	13.33x	20.00u	46.66n	31.027 d
Varietal means	**54.584 a**				**52.705 b**				**47.131 c**				**43.046 d**				**49.366**

LSD ($p<0.01$) values for salinity (S), varieties (V), and time (T) were 0.169 each, and for interactions (S x V x T)) were 0.675
Mean values followed by the same letter(s) within right side column (salinity) and bottom row (varieties) of the Table are not significantly different ($p<0.01$) using
LSD test. Similarly, values followed by the same letter(s) within column and rows (salinity treatments x varieties) in the center of the Table are not significantly different from each other

Table 6. Effect of salinity on lipid immobilization of four varieties of sunflower (*Helianthus annuus* L.) at three different intervals time (hours).

Salinity Treatments (S)	DO-728			DO-730			Hysun-33			Suncross-843			Salinity means
	48 h	96 h	144 h	48 h	96 h	144 h	48 h	96 h	144 h	48 h	96 h	144 h	
S_1	32.550 lm	27.327 rs	15.460 wx	28.500 qr	19.953 u	13.673 x	37.487 gh	28.473 qr	17.613 vw	39.957 ef	28.733 qr	19.257 uv	25.749 d
S_2	33.853 jkl	29.757 opq	25.687 st	29.453 pqr	23.507 t	17.233 vw	40.270 def	32.113 lmn	21.007 u	44.997 c	37.197 gh	32.007 lmno	30.590 c
S_3	34.883 ijk	32.203 lmn	28.710 qr	32.867 klm	30.053 nopq	21.067 u	44.953 c	38.653 fg	27.883 qrs	49.643 b	42.323 d	38.807 fg	35.171 b
S_4	36.030 hij	34.073 ijkl	31.547 mnop	36.293 hi	29.227 qr	24.097 t	49.787 ab	42.347	36.057 hij	52.010 a	49.893 ab	42.053 de	38.618 a
Varietal means	**30.173 c**			**25.494 d**			**34.720 b**			**39.740 a**			**32.532**

LSD ($p<0.01$) values for salinity (S), varieties (V), time (T) and for interactions (S x V x T)) are 0.65, 0.65, 0.57 and 2.27, respectively
Mean values followed by the same letter(s) within right side column (salinity) and bottom row (varieties) of the Table are not significantly different ($p<0.01$) using
LSD test. Similarly, values followed by the same letter(s) within column and rows (salinity treatments x varieties) in the center of the Table are not significantly different from each other

Results further exhibited that under salt stress, sunflower cultivars also responded significantly. A maximum lipid mobilization (25.49%) is recorded for cultivar DO-730 and minimum (39.74%) for Suncross-843. While the cultivars DO-728 and Hysun-33 is intermediate in lipid mobilization response toward salt stress, respectively (Table 6). Research studies revealed that there are many strategies to overcome the negative effects of salt stress. A good one strategy is the selection of cultivars and species for salinity tolerance. Similar cultivar variation under salt stress is also obtained in lentil crop (Ashraf *et al.,* 1992). The results of pair-wise comparison test of two factor interaction means *viz.,* SxT; SxT and VxT also showed that under each pair of variables a maximum significant lipid mobilization is recorded for non-stressed treatments (20.71%) variety DO-730 (19.02%) and time 144 h (16.50%). Whereas reverse is true as salinity level increases (Table 7abc).

Results further exhibited that as the concentration of applied salt increases; lipid mobilization inversely decreases when compared with their respective lipid contents (before sowing). This reduction is much pronounced in highest dose of applied salts (22.38 mS/cm) as compared to control (1.19 mS/cm) or even with their actual level of lipids present in seeds before soaking or sowing (Fig. 1). Cultivars are also responding differentially towards salt stress. Maximum reduction (14.85%) in mobilized lipids is noted for DO-728 and minimum for DO-730. Therefore, in term of lipid mobilization, cultivar DO-730 could be ranked as salt tolerant and DO-728 as salt sensitive. While the cultivar Hysun-33 and Suncross-843 is rated as salt intermediate in response respectively.

Conclusions

It can be concluded that as the concentration of salt increases there was a significant linear decline in imbibition, germination and lipid mobilization. A maximum decline in imbibition, germination and lipid mobilization is recorded in highest dose of salinity (22.38 mS/cm). The varietals response was also found to be highly significant. In term of imbibition and germination percentage, the maximum reduction is noted for Suncross-843 and minimum for DO-728. While in term of lipid mobilization, the maximum amount of immobilized lipid contents are recorded for Suncross-843 and minimum for DO-730. Therefore, in term of imbibitions and germination, DO-728 could be ranked as salt tolerant**,** but in term of lipid mobilization DO-730 is the salt tolerant cultivar.

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Table 7a. LSD of all pair-wise comparisons test for two factor interaction means of salinity and varieties (S x V) of various traits (imbibition, germination, lipid mobilization) of sunflower (*Helianthus annuus* L.) under salt stress.

Salinity levels	Varieties	Imbibition (%)	Germination (%)	Lipid mobilization
S_1	V1 = DO-728	22.912 a	70.837 b	25.112 i
	V2 = DO-730	20.722 b	72.495 a	20.709 k
	V3 = Hysun-33	19.490 c	67.938 c	27.858 h
	V4 = Suncross-843	18.452 de	67.090 d	29.316 g
S_2	V1 = DO-728	19.088 cd	57.507 e	29.766 g
	V2 = DO-730	17.800 e	55.825 f	23.398 j
	V3 = Hysun-33	16.907 f	49.997 g	31.130 ef
	V4 = Suncross-843	15.463 h	50.052 g	38.067 c
S_3	V1 = DO-728	16.222 g	49.162 h	31.392 e
	V2 = DO-730	15.747 gh	48.329 i	27.996 h
	V3 = Hysun-33	16.040 gh	41.479 j	37.163 c
	V4 = Suncross-843	13.753 ij	35.044 l	43.591 b
S_4	V1 = DO-728	13.577 ij	40.829 k	33.883 d
	V2 = DO-730	14.035 i	34.170 m	29.872 fg
	V3 = Hysun-33	13.318 j	29.108 n	42.730 b
	V4 = Suncross-843	10.592 k	20.000 o	47.986 a
LSD (p<0.01)		**0.682**	**0.338**	**0.654**

Mean values followed by the same letter(s) within a column are not significantly different (p<0.05) using LSD test

Table 7b. LSD of all pair-wise comparisons test for two factor interaction means of variety and time (V x T) of various traits (imbibition, germination, lipid mobilization) of sunflower (*Helianthus annuus* L.) under salt stress.

Varieties	Imbibition (%)	Germination (%)	Lipid mobilization
V1 = DO-728	13.478 f	21.677 l	34.329 d
	22.421 a	45.828 i	30.840 f
		72.498 c	25.351 g
		78.732 a	
V2 = DO-730	14.570 e	18.331 m	31.778 f
	19.582 b	47.491 h	25.685 g
		66.666 e	19.017 h
		78.332 a	
V3 = Hysun-33	13.793 f	15.255 n	43.124 b
	19.084 c	37.490 j	35.397 d
		61.698 f	25.640 g
		74.080 b	
V4 = Suncross-843	11.557 g	14.588 o	46.652 a
	17.773 d	32.500 k	39.537 c
		55.025 g	33.031 e
		70.073 d	
LSD (p<0.01)	**0.482**	**0.338**	**1.135**

Mean values followed by the same letter(s) within a column are not significantly different (p<0.05) using LSD test

Table 7c. LSD of all pair-wise comparisons test for two factor interaction means of salinity and time (S x T) of various traits (imbibition, germination, lipid mobilization) of sunflower (*Helianthus annuus* L.) under salt stress.

Salinity levels	Imbibition (%)	Germination (%)	Lipid mobilization
S_1	16.793 d	27.50 l	34.623 f
	23.994 a	53.33 h	26.122 j
		97.53 b	16.501 l
		100.00	
S_2	14.342 f	23.34 n	37.143 d
	20.288 b	48.33 i	30.643 h
		64.22 e	23.983 k
		77.50 c	
S_3	12.224 g	16.50 o	40.587 b
	18.657 c	36.66 k	35.808 e
		54.14 g	29.117 i
		66.71 d	
S_4	10.039 h	2.51 p	43.530 a
	15.722 e	24.99 m	38.885 c
		40.00 j	33.483 g
		56.61 f	
LSD (p<0.01)	**0.482**	**0.338**	**1.135**

Mean values followed by the same letter(s) within a column are not significantly different (p<0.05) using LSD test

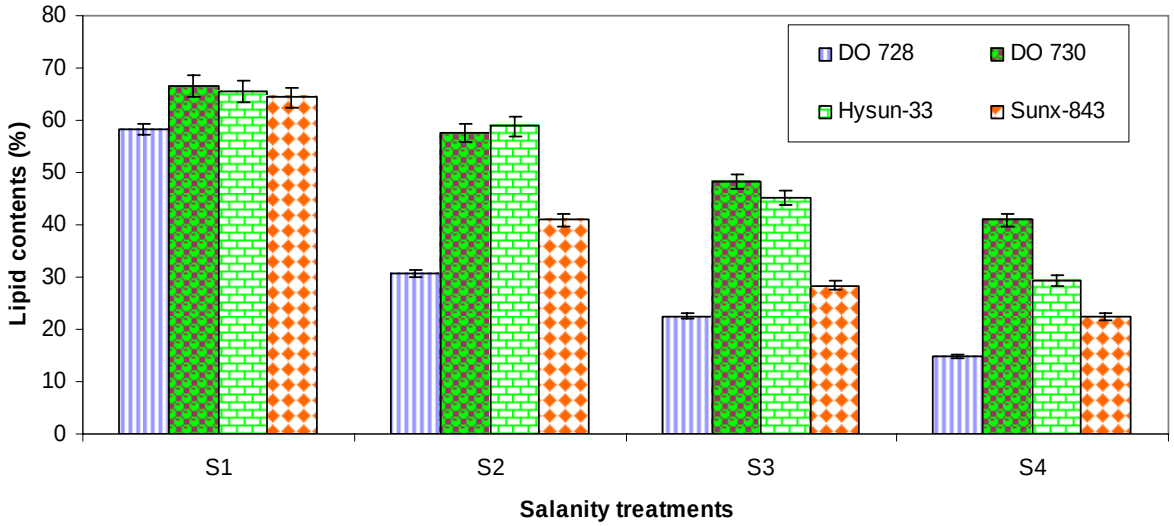

Fig. 1. Effect of salinity on the mobilized lipid contents (%) of four sunflower cultivars (after 144 h sowing) as compared with their respective lipid contents before sowing.

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