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Research Article

Morpho-physiological studies on moisture deficit stress tolerance in $F_{2:3}$ population of muskmelon (*Cucumis melo* L.)

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Abstract

Moisture deficit stress is a major abiotic factor affecting muskmelon quality and productivity globally. Here we have examined the response of morpho-physiological traits under moisture deficit stress condition in muskmelon parental lines *viz*., Durgapur Madhu (DM) and BS25 along with $F_{2:3}$ population derived from their cross for contrasting stress tolerance traits. Moisture stress reduced relative water content significantly ($p < 0.01$) with variation recorded among population lines. We observed that relative water content showed significant positive correlation ($r = 0.34$) with proline. A significantly increased value of the proline content (8.68 µg/gm), membrane stability index (27.11 mS/cm) and leaf area (24.43 cm^2) were observed in BS25 compared to DM genotype. Some population lines such as L8, L10, L83, L16, L21, L54, and L63 showed significantly higher values for proline content and relative water content in moisture stress condition. We observed variability for different traits in the $F_{2:3}$ population that allowed selection of individual lines with possibly greater potential of moisture stress tolerance, which can be useful in future breeding programs in muskmelon.

Keywords

moisture stress, proline, relative water content, chlorophyll

Introduction

Muskmelon (*Cucumis melo* L.) is an economically important summer season crop (2n=2x=24) of Cucurbitaceae family, cultivated preferably in hot climate of tropical and subtropical regions worldwide (Kirkbride, 1993). Variability for taste, nutrient composition and shape of melon fruit were already reported (Burger *et al.,* 2006; Fergany *et al.,* 2011). It exhibits excellent antioxidant potential and is used in the treatment for diabetes and respiratory problems (Kaur and Arora, 2011; Milind and Kulwant, 2011). China is the largest producer with 14.33 million tons and India stands at fifth position with 0.70 million tons as annual production of melon worldwide (FAOSTAT, 2013).

Drought is the major environmental stress that causes significant losses in productivity and crop development (Cattivelli *et al.,* 2008). In moisture deficit stress condition (MS), physiological and biochemical changes including functional losses were observed (Fini *et al.,* 2013). Researchers have examined number of biochemical and morphological responses of plants to moisture deficit stress such as altered relative water content, proline accumulation, chlorophyll content, membrane stability index and yield (Fabeiro *et al.,* 2002; Rizvi *et al.,* 2014; Liu *et al.,* 2015; Pandey *et al.,* 2016; Sachdeva *et al.,* 2017; Tiwari *et al.,* 2018). Knowledge on the physiological mechanisms that enables adaptation to MS and retaining productivity and development by overcoming stress could be helpful in screening of tolerant genotypes for its use in future programs of molecular breeding (Zaharieva *et al.,* 2001).

In the recent years, most studies were based on the morphological parameters (plant and fruit) in drought conditions (Kusvuran, 2012; Ibrahim, 2012; Mirabad *et al.,* 2013; Patil *et al.,* 2014; Mundalia *et al.,* 2015; Pandey *et al.,* 2016). The phenotypic and genotypic correlation coefficient was also studied for fruit yield and its related traits in muskmelon under MS conditions (Mishra *et al.,* 2016; 2017). Melon production and productivity improve with selection of tolerant genotypes for moisture stress (Rashidi and Seyfi, 2007). Although, a number of drought tolerant varieties of other crops have been reported earlier (Mir *et al.,* 2012), but only limited data is accessible on moisture deficit stress tolerant in melon varieties and population (Cabello *et al.,* 2009). The present study details on the morpho-physiological variability for growth and association between different traits under well watered and moisture deficit stress conditions in muskmelon cultivars and their population. The information derived from the study would be useful in future breeding programs of melon for moisture deficit stress tolerance.

Materials and Methods

The present experiment was carried out during summer season (April-July) of 2016 at ICAR-National Bureau of Plant Genetic Resources (NBPGR), New Delhi, India. The population $F_{2:3}$ were developed by crossing muskmelon inbreds *viz.*, Durgapur Madhu (DM) (female) genotype being susceptible to MS with the tolerant genotype BS25 (male). The parents were selected based on their contrasting response to moisture stress tolerance (Pandey *et al.*, 2013; 2016). The number of fruits and total soluble sugar (TSS) per cent were higher in DM along with best response for general combining ability effect (Randhawa and Singh, 1990; Choudhary *et al.*, 2006). Productivity of BS25 was quite similar in both the conditions (well watered and moisture stress) and its drought tolerant efficiency per cent (DTE %) was higher than other genotypes as calculated through field experiments (Pandey *et al.*, 2013; 2016). The F_1 between DM × BS25 was subsequently self-pollinated to produce 96 F_2 progenies and $F_{2:3}$ families. A total of 86 $F_{2:3}$ families were evaluated during the summer season along with the parents and F_1 hybrid. Field experiment to evaluate the population for MS tolerance was performed in a randomized block design with five plants in each row with three replications under two treatment conditions *viz.*, well-watered (WW) and moisture deficit stress (MS). Standard agronomic procedure was followed during crop stand for both treatment conditions. Plants were irrigated once in ten days until unless specified. Thirty days after sowing, irrigation was stopped in one set (MS) while it is continued (once in ten days) in the other set (WW) as per requirement and observation was noted. After exposure to thirty days moisture stress, the experimental set was re-irrigated for two times at five days interval and recovery of the plants (61–70 days after germination) were observed.

Morpho-Physiological observations were recorded for parents, F_1 and $F_{2:3}$ families. A calibrated Gopher soil moisture profiler (Soil moisture technology, Gopher, Australia) was used to measure soil moisture content at 5-10 days interval. The soil moisture percentage was also recorded before and after irrigation. Measured soil moisture percentage was averaged of three readings between plant row and middle of two lines (Kang *et al.*, 2002). Three healthy leaves were collected from three different plants randomly for calculating the relative water content (RWC) (Barrs and Weatherley, 1962). Membrane stability index (MSI) was calculated by noting the electrical conductivity of leaf samples in double distilled water at 40°C and 100°C according to Sairam (1994).

Three readings of chlorophyll content of flag leaf were measured using chlorophyll meter (SPAD-502, Soil Plant Analysis Development section, Japan). The average values were used for analysis as SPAD units (Martinez and Guiamet, 2004). Leaf area was also recorded using portable laser area meter (C1-203, CID Bio-science, USA) (Yin *et al.*, 2005). Proline was estimated using ninhydrin method reported by Bates et al. (1973). Protein content was measured using bovine serum albumin (BSA) as a standard (Bradford, 1976) at 595 nm wavelength, using a spectrophotometer (DU 640, Beckman Instruments Inc., USA).

Factorial ANOVA was carried out for the observed morpho-physiological traits in WW and MS conditions of melon population to study its significance statistically (Rangaswamy, 2010). The correlation coefficient matrix was also analyzed using Genstat software (v.18.1) for the traits observed during this study.

Results and Discussion

Soil moisture of experimental farm was recorded to be 70-80 per cent and 7-9 per cent respectively at field capacity and at wilting point using the Gopher soil moisture profiler. Under drought conditions soil moisture was observed to be 10-20 per cent between 35-75 days after sowing. Lower relative water content was observed in the MS (50%) plant compared to the WW (84%) plants. Statistical analyses indicated that RWC values between treatments and among the individual lines including parents (Table 1) were significant ($p < 0.01$). Therefore, genetic variability might exist between population lines for key traits. A range of traits was found in the $F_{2:3}$ population under moisture stress and well watered conditions. The parental line had RWC values under moisture stress condition (DM mean, 59.18%; s.d, 0.37; BS25 mean, 73.69%; s.d, 0.12) and under well watered conditions (DM mean, 76.09%; s.d, 0.47; BS25 mean, 74.22%; s.d, 0.31). In addition, several lines of $F_{2:3}$ population having certain trait values greater as well as lesser than the parental lines were identified in the stress conditions. There were individual lines that had high RWC under moisture stress, such as L83 (MS: 70.58%; WW: 80.80%) and L8 (MS: 71.49%; WW: 76.48%) whereas other lines such as L79 (MS: 41.79%; WW: 75.70%) and L21 (MS: 42.52%; WW: 87.36%) showed low RWC (Supplementary Table S1).

Under the moisture stress conditions electric conductivity of the leaf was lesser in comparison to well watered conditions. Average MSI declined in the moisture deficit field throughout stress period with statistically significant difference observed among the lines ($p < 0.01$) and between treatments. Some lines showed high MSI in MS condition such as L17 (MS: 80.61mS/cm; WW: 90.91 mS/cm) and L16 (MS: 70.08 mS/cm; WW: 84.53 mS/cm) whereas other lines showed low MSI, L58 (MS: 1.20 mS/cm; WW: 20.68 mS/cm) and L50 (MS: 1.51 mS/cm; WW: 26.82 mS/cm). A range in traits was observed in the $F_{2:3}$ populations 1.20−90.61 mS/cm (DM mean, 8.37 mS/cm; s.d, 0.04; BS25 mean, 19.10 mS/cm; s.d, 0.40) in MS condition. While under well watered condition traits values 2.64−75.15 mS/cm were observed (DM mean, 25.49 mS/cm; s.d, 1.03; BS25 mean, 27.72 mS/cm; s.d, 0.82).

Accumulation of proline content in the plants under MS treatment was observed. A significant difference was observed among the individual lines for proline ($p < 0.01$) and between the treatments as well (Table 2). Proline content in the $F_{2:3}$ population ranged between 2.42−9.93 µg/gm in MS condition (DM mean, 4.82 µg/gm; s.d, 0.04; BS25 mean, 8.67 µg/gm; s.d, 0.006) and 2.29−5.61 µg/gm in WW condition (DM mean, 4.01 µg/gm; s.d, 0.007; BS25 mean, 2.24 µg/gm; s.d, 0.004). Some individual lines were recorded with high proline in MS situations, such as L54 (MS: 9.92 µg/gm; WW: 2.79 µg/gm) and L63 (MS: 7.46 µg/gm; WW: 5.61 µg/gm) whereas other lines showed low proline [L28 (MS: 2.42 µg/gm; WW: 2.35 µg/gm) and L51 (MS: 2.46 µg/gm; WW: 2.35 µg/gm)]. In the $F_{2:3}$ populations some lines showed higher and lower values of proline contents under drought stress condition in comparison to parental lines.

Genotypes among the $F_{2:3}$ families and the parents showed significant differences ($p < 0.01$) in protein content and between treatments (WW and MS) as well. Protein content ranged between 0.34 and 4.76 µg/ml under MS condition (DM mean, 4.11 µg/ml; s.d, 0.053; BS25 mean, 4.30 µg/ml; s.d, 0.729) and 2.29−4.79 µg/ml under WW conditions (DM mean, 4.62 µg/ml; s.d, 0.009; BS25 mean, 4.68 µg/ml; s.d, 0.010) in the $F_{2:3}$ population (Table 3). Reduction in mean value of protein was observed between two parents and population (Supplementary Table S2). Some individual lines showing high protein content under moisture stress were: L65 (MS: 4.763 µg/ml; WW: 4.103 µg/ml) and L81 (MS: 4.683 µg/ml; WW: 2.468 µg/ml). Whereas, other lines such as L39 (MS: 0.339 µg/ml; WW: 4.511 µg/ml) and L17 (MS: 0.615 µg/ml; WW: 2.948 µg/ml) were observed with low proline content.

There was no significant difference between the plants studied in MS and WW conditions. Chlorophyll content was moderately influenced by stress treatment. Reduction in the leaf area under moisture stress was observed when compared to well watered plants. ANOVA showed significant differences among the individual lines for LA ($p < 0.01$) and between treatments (Supplementary Table S4). Variation in the mean value was assessed between two parents and population (Fig. 1). LA ranged between 10.35 and 55.61 cm^2 in moisture stress conditions (DM mean, 19.31 cm^2; s.d, 0.91; BS25 mean, 24.42 cm^2; s.d, 3.47) and 23.50−61 cm^2 under well watered conditions (DM mean, 26.99 cm^2; s.d, 3.15; BS25 mean, 28.70 cm^2; s.d, 2.52). There were some lines that showed large leaf area (Supplementary Table S2) under MS conditions.

Under correlation matrix, we observed RWC showing positive significant correlation with protein content, leaf area and MSI (r = 0.348, 0.196 and 0.176 respectively) (Table 4). It showed a negative significant correlation with proline. Leaf area showed positive significant correlation with MSI, and protein. A negative significant correlation between proline and MSI was also observed.

In the present study, the variation in the traits observed under moisture stress treatment (MS) indicated that selection using some of these drought related characteristics could be effective in developing tolerant cultivars. However, the efficiency of selection in a population basically relies on heritability. Variation between the individual population lines and parental lines were identified in stress condition, suggesting possible transgressive segregation in the population and indicates the population is normally distributed (Chai *et al.,* 2016). RWC and MSI are important criteria reflecting water status or potential for selection of drought tolerant cultivars (Valentovic *et al.,* 2006; Rad *et al.,* 2013). The RWC percentage is significantly decreased during drought stress (Liu *et al.,* 2015). MSI is a measure of membrane integrity that differentiates the tolerant and susceptible genotypes (Surendar *et al.,* 2013). In the present study, a significant difference in RWC and MSI values was observed in melon parents and their progenies. The Parental genotype BS25 showed higher percentage of RWC and MSI in comparison to DM under stress and well watered condition which shows its tolerance efficiency

(Supplementary Table S1). The population lines also exhibit lower and higher values in treatments. The differences in RWC and MSI between genotypes tolerant and susceptible to moisture stress recorded in this study are in line with earlier reports (Pandey *et al.,* 2016, Ansari *et al.,* 2017). The electrolyte leakage of the susceptible maize genotype increased in comparison to tolerant (Valentovic *et al.*, 2006). In similar earlier studies in wheat (Sairam and Srivastava, 2001, Liu et al. 2015), muskmelon (Kusvuran, 2012) and chickpea (Sachdeva *et al.,* 2017), significant differences were observed in RWC and MSI between tolerant and susceptible genotypes.

Under moisture stress condition, reduction in leaf area was observed in comparison to WW condition. The results obtained here are similar to results revealed in cantaloupe (Cabello *et al.,* 2009). Decrease in leaf area might associate with water reduction in active root part that in turn interrupts plant development by disturbing physiological processes (Songsri *et al.,* 2009).

The osmolyte proline accumulation enables plant to adapt under drought (Kusvuran and Dasgan, 2017). Proline content significantly increased in parental line BS25 and variation in population lines under water stress conditions was observed (Supplementary Table S3). Earlier findings in various crops support the increased proline content under stress conditions such as mulberry (Ramanjulu and Sudhakar, 2000), maize (Moussa and Abdel-Aziz, 2008), mothbean (Sachdeva *et al.,* 2016), common bean (Kusvuran and Dasgan, 2017) and muskmelon (Ansari *et al.,* 2017). It helps in regulation of cellular redox status by maintaining the protein and membrane structure in drought stress (Yamada *et al.,* 2005). The decrease in protein content during stress was also reported in other crops like groundnut (Hui Fang and Xiao Ping, 2004), sesame (Fazeli *et al.*, 2007) and maize (Mohammadkhani and Heidari, 2008). Water stress causes reduction in carbon skeleton necessary for amino acid and leads to decrease in photosynthesis as well as synthesis of proteins (Mohammadkhani and Heidari, 2008).

Under moisture stress conditions parents and segregating population showed earlier male flower growth than well watered plants. DM genotype and some population lines showed hermaphroditism during the flowering in both conditions (MS and WW). Maintenance of water status in plants is thus considered as a mechanism for moisture stress tolerance in BS25 genotype. A character by character examination showed that different characters were differentially associated with each other (Taha *et al.* 2003). The correlation of RWC was positively significant with protein, MSI and leaf area. The correlation of chlorophyll content was insignificant with all of the traits studied. Correlation coefficients revealed that different traits were differentially associated with each other (Sharma *et al.,* 2014). Significant correlation variation was reported for morphological, physiological, yield components and yield in muskmelon (Tomar *et al.,* 2008; Reddy *et al.,* 2013; Mishra *et al.,* 2016; 2017).

Relative water content, membrane stability index and proline can be used for selection of moisture stress tolerant genotypes. Leaf area and protein content are also important traits for selection under drought stress. High values of traits variation in population could be helpful for selection of superior muskmelon lines for retaining yield in water stress condition. The ability of plants to maintain water status in moisture stress helps them to sustain during the period of drought and recovery after irrigation (Ansari *et al.,* 2017). The combined responses of the $F_{2:3}$ populations that performed better under moisture deficit conditions can potentially be considered for future breeding programs. For example, L54 and L63 had high proline content (9.92 and 7.46 µg/gm), moderate RWC (47.7 and 58.4%), protein (4.65 and 4.41 µg/ml), chlorophyll (34.3 and 36.4 SPAD unit), leaf area (40.4 and 30.9cm^2) and low MSI (2.69 and 20.34 mS/cm). The final selection of the line is very complex due to combined effects of the RWC and MSI both related to leaf area. The segregating population also showed the combined variation between proline, protein and chlorophyll content under moisture deficit conditions. Differences between the parental lines during the moisture stress for many traits indicate that some of the adaptation mechanisms to moisture deficit stress might be not-identical in the two genotypes. So, it could be mapped and selected for different trait combinations in the offspring for future work in breeding and research.

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Table 1. Analysis of Variance for relative water content and membrane stability index traits in melon genotypes

Source of var.	**DF**	**Relative water content**			**Membrane stability index**		
		SS	***MS***	***F-value***	***SS***	***MS***	***F-value***
Replication	2	7.65	3.82	9.19**	0.89	0.44 ^{ns}	0.91 ^{ns}
Genotype	85	20408.17	240.09**	576.86**	108711.5	1278.9**	2618.6**
Drought	1	23976.42	23976.42**	57606**	9130.76	9130.7**	18694**
G × D	85	18039.32	212.22**	509.90**	101662.2	1196**	2448**
Trt SS	171	62423.92	365.05**	877.08**	219504.5	1283.6**	2628.2**
Error	342	142.34	0.41		167.03	0.48	
Total	515	62573.91			219672.4		

ns = non significant, SS = sum of square, MS = mean square
DF = degree of freedom, *significant at ($p \leq 0.05$) , **significant at ($p \leq 0.01$)

Table 2. Analysis of Variance for proline and protein content in melon genotypes

Source of var.	**DF**	**Proline content**			**Protein content**		
		SS	**MS**	***F-value***	**SS**	**MS**	***F-value***
Replication	2	0.006	0.003	3.43*	0.06	0.03	4.44*
Genotype	85	403.80	4.75**	4708**	103.57	1.21**	168.1**
Drought	1	71.40	71.40**	70777**	51.00	51.00**	7039**
G × D	85	167.37	1.96**	1951**	82.69	0.97**	134.4**
Trt SS	171	642.58	3.75**	3724**	237.28	1.38**	191.4**
Error	342	0.34	0.001		2.47	0.007	
Total	515	642.93			239.82		

ns = non significant, SS = sum of square, MS = mean square
DF = degree of freedom, *significant at ($p \leq 0.05$) , **significant at ($p \leq 0.01$)

Table 3. Variation for morpho-physiological traits in melon parents and $F_{2:3}$ population under well water (WW) and moisture deficit stress (MS) conditions

Trait	Treatment	Mean	SD	MIN	MAX	CV	Skewness	Kurtosis	Confidence level	DM		BS25	
										MIN	MAX	MIN	MAX
RWC	WW	74.59	9.05	48.29	96.47	0.121	-0.478	0.299	1.912	75.55	76.94	73.94	74.56
	MS	60.95	8.30	41.79	80.59	0.136	-0.042	-0.459	1.754	58.85	59.58	73.57	73.82
MSI	WW	34.16	16.69	2.64	75.15	0.488	0.242	-0.351	3.528	24.65	26.65	19.05	20.65
	MS	25.81	23.28	1.20	90.61	0.902	0.915	-0.140	4.921	8.33	8.42	26.75	27.55
Chl	WW	34.28	4.42	24.47	43.10	0.128	-0.168	-0.506	0.933	35.80	38.10	34.90	42.40
	MS	34.89	2.61	29.37	41.23	0.074	-0.026	-0.233	0.550	32.70	34.10	29.00	32.50
LA	WW	38.44	7.15	23.50	61.00	0.186	0.232	0.466	1.511	25.09	30.64	26.68	31.54
	MS	31.68	9.66	10.35	55.61	0.305	0.312	-0.332	2.042	18.34	20.15	21.48	28.26
Proline	WW	3.09	0.65	2.29	5.61	0.211	1.495	2.873	0.138	4.01	4.02	2.54	2.55
	MS	3.84	1.35	2.42	9.93	0.350	2.079	5.794	0.284	4.78	4.87	8.68	8.69
Protein	WW	4.41	0.39	2.29	4.79	0.088	-2.807	10.765	0.082	4.62	4.64	4.67	4.69
	MS	3.78	0.76	0.34	4.76	0.201	-2.178	6.895	0.161	4.08	4.18	3.53	4.98

Standard deviation (SD), Coefficient of variation (CV), (relative water content (RWC), membrane stability index (MSI), chlorophyll (Chl), leaf area (LA)

Table 4. Correlation coefficient among morpho-physiological traits in melon genotypes

Traits	Chlorophyll	LA	MSI	Protein	Proline
LA	0.039				
MSI	0.005	0.189**			
Protein	0.002	0.227**	0.116		
Proline	0.04	-0.114	-0.153**	-0.079	
RWC	-0.083	0.196**	0.176**	0.348**	-0.31**

*significant at $p \leq 0.05$, **significant at $p \leq 0.01$

Relative water content (RWC), membrane stability index (MSI), leaf area (LA)

Supplementary Table S1. Data showing variation in mean value of three replicates ± standard error for relative water content (RWC) (%), membrane stability index (MSI) (mS/cm) and chlorophyll content (Chl) (SPAD unit) of melon parents and $F_{2:3}$ population lines in well watered (WW) and moisture deficit stress (MS) conditions.

Lines	RWC (WW)	RWC (MS)	MSI (WW)	MSI (MS)	Chl (WW)	Chl (MS)
DM	76.09±0.27	59.18±0.21	25.49±0.59	8.37±0.02	36.9±0.66	33.33±0.41
BS25	74.22±0.17	73.69±0.07	27.72±0.47	19.10±0.23	38.06±2.24	31.13±1.08
F1	68.69±0.55	50.58±0.24	32.77±0.36	21.53±0.23	35.36±2.60	34.9±1.36
L1	71.45±0.25	70.63±0.58	68.34±0.59	40.16±0.55	30.96±0.08	24.6±3.21
L2	85.31±0.34	56.52±0.17	68.81±0.63	46.75±0.32	36.36±3.03	34±1.32
L3	83.70±0.29	61.31±0.23	5.38±0.36	4.45±0.55	34.76±3.03	26.36±1.60
L4	76.40±0.44	68.49±0.19	43.30±0.31	39.10±0.21	31.06±2.04	30.26±2.88
L5	84.92±0.46	60.59±0.13	25.58±0.36	10.14±0.17	37.26±0.23	34.43±4.61
L6	64.58±0.56	62.25±0.43	46.26±0.23	29.87±0.19	36.3±4.47	32.33±0.77
L7	74.54±0.21	67.19±0.34	43.32±0.34	39.44±0.38	35.2±0.25	34±1.56
L8	76.48±0.27	71.49±0.20	9.80±0.59	4.60±0.25	33.96±0.99	31.76±4.34
L9	79.07±0.20	60.50±0.13	5.51±0.31	1.79±0.19	36.13±1.19	33.63±4.79
L10	85.46±0.44	72.57±0.21	36.43±0.29	34.22±0.67	34.53±1.53	32.63±3.18
L11	84.52±0.22	60.51±0.23	45.33±0.28	38.94±0.63	35.53±0.69	32.16±4.37
L12	96.46±0.56	61.98±0.49	75.15±0.19	33.86±0.42	40.96±0.72	37.56±2.09
L13	80.54±0.57	55.02±0.11	70.32±0.62	50.36±0.37	37.7±0.78	32.8±3.50
L14	83.80±0.04	61.54±0.15	65.64±0.13	60.42±0.30	36.36±1.16	35.83±1.65
L15	80.92±0.91	60.72±0.27	61.33±0.30	57.05±0.07	39.96±1.94	33.26±2.75
L16	75.15±0.84	62.42±0.26	84.53±0.19	70.08±0.23	41.53±2.33	36.56±2.24
L17	71.41±0.22	52.68±0.55	90.91±0.14	80.61±0.34	41.83±2.53	37.36±2.61
L18	77.36±0.36	65.30±0.40	28.11±0.19	1.79±0.22	37.4±1.66	33.8±2.15
L19	76.41±0.11	60.55±0.15	76.89±0.32	33.59±0.35	34.4±0.87	30.13±2.74
L20	76.32±0.35	58.41±0.21	40.23±0.21	38.02±0.27	36.66±0.29	31.56±3.27
L21	87.36±0.20	42.52±0.16	44.26±0.28	23.41±0.24	38.8±1.74	31.4±2.50
L22	61.35±0.35	61.54±0.17	71.14±0.14	18.51±0.38	35.53±2.25	30.8±0.37
L23	74.73±0.63	68.30±0.29	34.13±0.20	9.06±0.40	40±0.45	34.33±0.53
L24	87.34±0.38	70.55±0.17	58.49±0.18	48.29±0.27	43.03±1.75	35.73±0.43
L25	66.69±0.23	63.21±0.18	42.75±0.25	16.38±0.27	42.1±1.87	34.8±0.46
L26	61.29±0.33	48.33±0.24	32.40±0.23	3.10±0.45	43.1±1.62	39.73±1.42
L27	63.75±0.25	62.47±0.24	44.41±0.22	13.34±0.26	36.2±1.12	34.33±1.44
L28	69.20±0.31	59.46±0.24	42.19±0.21	31.29±0.33	38.1±0.45	36.66±0.99
L29	80.58±0.64	58.23±0.33	53.55±0.14	6.43±0.49	35.96±1.41	30.83±0.83
L30	75.49±0.18	66.31±0.27	62.36±0.37	28.08±0.33	36.2±1.34	28.7±1.80
L31	77.38±0.86	66.53±0.24	63.34±0.12	7.60±0.69	35.2±1.25	28.06±1.93
L32	76.58±0.31	60.44±0.20	48.65±0.33	20.39±0.17	37.36±0.37	36±0.05
L33	69.44±0.59	65.57±0.05	22.11±0.38	20.01±1.01	35.1±0.36	34.66±0.81
L34	68.40±0.41	66.37±0.60	57.44±0.23	23.99±0.37	38.73±1.28	34.36±1.33
L35	86.52±0.11	46.88±0.65	43.55±0.36	10.87±0.52	40.06±1.08	37.03±0.44
L36	70.87±0.16	52.40±0.39	69.36±0.26	62.39±0.12	35.53±0.92	33.2±0.70
L37	82.63±0.73	64.56±0.18	65.53±0.17	6.45±0.13	36.7±0.56	32.06±1.87
L38	76.41±0.23	72.57±0.08	41.25±0.30	39.68±0.18	37.5±0.73	25.66±0.56
L39	80.13±0.86	55.66±0.50	45.77±0.48	17.29±0.35	37.9±2.42	24.4±0.60
L40	76.28±0.37	70.59±0.16	30.39±0.58	14.49±0.28	29.23±1.26	27.93±2.94
L41	70.13±0.92	60.71±0.26	13.39±0.63	3.64±0.39	39.33±0.81	36.66±0.57
L42	63.90±0.12	51.43±0.27	50.75±0.05	23.43±0.35	36.63±3.03	31.06±0.20
L43	86.46±0.60	63.66±0.01	16.65±0.17	1.58±0.25	33.66±1.10	30.7±0.92
L44	79.43±0.12	70.51±0.12	12.27±0.43	2.33±0.34	34.76±3.03	28±1.13
L45	72.65±0.57	47.61±0.13	21.09±0.14	17.30±0.34	31±1.79	30.56±1.51
L46	64.68±0.09	54.63±0.04	39.59±0.18	33.64±0.12	41.5±0.66	35.23±1.81
L47	69.18±0.28	53.39±0.49	41.40±0.63	2.64±0.23	35.1±2.34	32.6±1.27
L48	77.35±0.18	57.47±0.24	32.84±0.35	25.03±0.08	35.46±4.42	24.33±0.32

L49	71.94±0.33	74.60±0.24	48.46±0.54	14.37±0.27	32.4±1.22	30.26±0.55
L50	78.36±0.23	53.46±0.16	26.82±0.16	1.51±0.21	35.5±1.28	29.5±0.36
L51	85.69±0.20	73.43±0.34	42.07±0.47	65.67±0.26	34.06±3.12	32.36±1.73
L52	67.97±0.17	65.55±0.06	38.46±0.56	1.34±0.07	35.36±1.40	30.93±1.26
L53	60.44±0.13	58.49±0.16	21.24±0.26	12.70±0.43	38.03±3.13	30.36±4.96
L54	54.88±0.74	47.73±0.09	17.47±0.57	2.69±0.08	34.43±2.10	31.3±4.99
L55	86.50±0.55	57.75±0.01	6.38±0.60	43.29±0.36	36.56±1.38	35.73±2.37
L56	65.61±0.22	52.81±0.46	41.54±0.09	2.66±0.10	36.96±1.50	35.96±1.08
L57	70.91±0.87	55.56±0.16	13.36±0.36	16.33±0.29	35.73±5.20	34.63±0.92
L58	83.13±0.20	69.38±0.05	20.68±0.49	1.20±0.16	37.83±0.89	33.56±0.21
L59	82.73±0.64	52.87±0.08	35.24±0.30	56.59±0.45	39.56±1.32	29.36±2.18
L60	77.58±0.22	68.77±0.16	49.32±0.56	6.66±0.33	37.76±1.38	35.5±1.59
L61	76.41±0.44	63.77±0.09	29.13±0.36	8.63±0.05	34.53±0.69	30.6±1.56
L62	63.58±0.51	51.55±0.21	33.94±0.12	4.53±0.26	34.9±1.87	32.03±1.78
L63	76.79±0.08	58.45±0.22	16.19±0.23	20.34±0.19	39.63±0.64	36.43±0.60
L64	69.29±0.20	64.51±0.22	16.70±0.74	2.85±0.18	35.13±0.49	34.26±1.31
L65	61.55±0.15	67.42±0.10	5.35±0.61	28.12±0.39	33.76±2.91	27.93±0.95
L66	52.22±0.84	47.36±0.26	40.67±0.66	2.53±0.28	35.73±4.31	28.7±1.02
L67	73.91±0.38	71.72±0.07	55.40±0.52	7.27±0.13	37.7±2.42	33.7±1.98
L68	73.48±0.23	53.56±0.79	40.50±0.55	10.22±0.21	31.53±5.41	30.23±2.16
L69	72.48±0.72	70.75±0.07	31.24±0.84	2.16±0.31	38.1±4.14	36.06±0.95
L70	74.49±0.21	53.68±0.10	25.01±0.87	44.25±0.32	40.53±1.06	31.86±1.60
L71	76.26±0.84	57.61±0.05	23.04±0.89	25.82±0.22	32.06±1.90	30.76±1.12
L72	83.70±0.13	51.42±0.17	44.30±0.76	15.06±0.51	37.23±2.32	35.4±0.96
L73	89.23±0.34	75.73±0.51	47.13±0.28	22.24±0.26	32.16±1.67	29.73±1.6
L74	76.49±0.24	56.49±0.13	44.25±0.30	8.58±0.17	37.2±6.05	31.76±0.18
L75	83.43±0.20	58.71±0.10	15.60±0.54	51.09±0.31	39.1±2.50	37.16±0.61
L76	84.01±0.45	62.95±0.84	55.54±0.57	18.62±0.05	40.96±3.09	32.23±0.67
L77	70.55±0.22	55.39±0.24	20.44±0.60	4.37±0.10	31.63±1.23	28.76±2.90
L78	85.35±0.86	46.61±0.50	30.89±0.88	6.60±0.03	36.7±3.78	34.5±2.19
L79	75.70±0.08	46.79±0.07	41.06±0.79	11.67±0.18	38.7±1.61	36.73±2.70
L80	72.25±0.34	67.17±0.19	51.87±0.87	36.40±0.13	38.33±3.52	31.83±1.68
L81	72.00±0.34	58.50±0.20	44.64±0.53	15.54±0.05	36.13±4.16	33.93±0.29
L82	60.32±0.33	55.41±0.39	30.49±0.47	8.18±0.24	38.2±0.60	33.9±2.17
L83	80.80±0.43	70.58±0.17	40.38±0.78	3.42±0.22	32.4±0.37	31.53±0.60

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Supplementary Table S2. Data showing mean value variation of three replicates ± standard error for leaf area (LA) (cm^2), proline content (µg/gm) and protein content (µg/ml) of melon parents and $F_{2:3}$ population lines in well watered (WW) and moisture deficit stress (MS) conditions.

Lines	LA (WW)	LA (MS)	Proline (WW)	Proline (MS)	Protein (WW)	Protein (MS)
DM	26.99±1.82	19.31±0.52	4.016±0.004	4.825±0.024	4.627±0.005	4.119±0.030
BS25	28.70±1.45	24.42±2.00	2.546±0.002	8.679±0.003	4.681±0.006	4.306±0.421
F1	32.45±0.64	23.17±1.16	2.751±0.003	3.041±0.005	4.542±0.005	4.052±0.016
L1	35.88±1.68	30.17±2.54	2.911±0.006	3.129±0.005	4.622±0.004	4.523±0.006
L2	36.07±3.23	33.44±0.61	3.152±0.003	3.931±0.038	3.748±0.007	3.616±0.001
L3	37.25±1.76	34.56±1.31	2.598±0.005	2.761±0.007	4.536±0.005	4.471±0.005
L4	33.09±2.80	32.55±1.48	2.847±0.004	3.050±0.005	4.085±0.025	4.036±0.001
L5	41.20±3.74	38.08±2.33	3.148±0.004	3.468±0.006	4.360±0.011	4.465±0.000
L6	54.50±3.76	35.26±2.55	2.596±0.005	3.100±0.005	4.132±0.015	3.941±0.001
L7	44.34±3.64	29.19±0.43	3.582±0.035	5.552±0.005	3.849±0.036	3.744±0.001
L8	41.32±1.87	40.68±2.50	2.514±0.004	2.664±0.117	4.605±0.016	4.620±0.001
L9	40.36±3.75	27.60±1.44	2.612±0.005	2.728±0.005	4.094±0.017	3.584±0.001
L10	41.17±2.21	32.41±2.17	3.120±0.004	3.918±0.007	4.451±0.006	4.459±0.002
L11	49.75±2.12	37.12±1.23	2.559±0.003	2.660±0.004	4.354±0.019	4.530±0.002
L12	47.97±1.92	35.45±2.77	3.956±0.033	5.757±0.035	4.724±0.005	4.505±0.004
L13	61.00±0.79	50.90±0.90	2.993±0.003	3.552±0.004	4.529±0.007	4.125±0.007
L14	48.44±2.75	31.43±3.14	2.829±0.004	3.060±0.006	4.631±0.005	4.294±0.003
L15	49.68±2.77	39.82±4.01	2.765±0.003	2.429±0.004	4.515±0.005	4.356±0.004
L16	41.58±4.32	36.44±2.94	3.220±0.049	6.284±0.039	4.682±0.009	4.228±0.003
L17	47.57±1.64	39.24±0.57	2.948±0.004	4.626±0.055	4.583±0.001	0.615±0.006
L18	55.57±3.73	52.61±3.19	3.128±0.006	3.265±0.005	4.423±0.005	4.385±0.007
L19	44.61±3.47	40.60±3.37	2.829±0.014	2.436±0.004	4.622±0.001	4.425±0.175
L20	46.37±2.53	35.79±3.23	2.924±0.007	3.623±0.010	4.246±0.001	3.956±0.017
L21	34.85±4.69	35.33±3.27	3.009±0.004	5.638±0.008	4.512±0.001	3.441±0.002
L22	42.04±1.82	39.54±2.17	2.555±0.005	2.613±0.006	4.676±0.065	4.086±0.014
L23	53.37±5.78	44.39±3.12	2.601±0.004	2.811±0.006	4.739±0.002	4.176±0.026
L24	41.39±1.34	35.82±6.45	3.124±0.007	3.798±0.037	4.719±0.002	4.0217±0.010
L25	41.33±4.38	16.50±0.73	2.937±0.005	3.073±0.012	3.983±0.001	3.109±0.032
L26	37.84±3.61	36.82±2.37	3.324±0.012	4.721±0.034	4.505±0.002	4.072±0.090
L27	36.34±3.07	29.50±2.35	2.327±0.005	4.658±0.009	4.385±0.009	3.831±0.064
L28	40.93±1.78	26.04±3.14	2.354±0.005	2.420±0.027	4.723±0.004	3.581±0.107
L29	41.74±1.03	39.27±4.22	3.138±0.002	3.494±0.008	2.293±0.001	4.301±0.111
L30	38.92±3.08	32.07±0.69	2.770±0.003	3.34±0.013	4.530±0.001	3.324±0.264
L31	29.51±2.54	26.94±1.31	2.353±0.002	2.525±0.008	4.675±0.004	4.294±0.105
L32	38.74±5.46	27.37±0.53	2.360±0.003	2.525±0.003	4.617±0.001	3.917±0.030
L33	48.40±2.95	38.49±1.68	2.838±0.006	3.810±0.004	4.517±0.001	3.917±0.004
L34	34.78±3.34	19.50±0.54	2.311±0.004	3.640±0.012	4.583±0.001	4.131±0.030
L35	45.45±1.60	27.72±2.16	3.570±0.011	4.660±0.025	4.679±0.001	4.446±0.113
L36	27.69±1.01	23.44±1.31	2.763±0.005	2.931±0.005	4.742±0.006	4.261±0.088
L37	43.60±5.48	30.39±2.71	2.305±0.004	4.149±0.007	4.622±0.005	4.276±0.098
L38	31.45±3.52	26.68±3.36	2.286±0.005	3.526±0.005	4.351±0.002	3.919±0.044
L39	39.70±5.16	32.61±0.80	2.706±0.002	2.883±0.044	4.511±0.006	0.338±0.010
L40	38.21±2.37	15.61±0.60	2.590±0.008	4.117±0.005	3.163±0.012	3.741±0.200

L41	34.20±1.84	21.18±0.93	2.722±0.004	2.921±0.005	4.372±0.010	3.938±0.024
L42	40.11±3.16	17.42±1.12	3.393±0.006	5.009±0.017	4.788±0.003	3.730±0.009
L43	42.30±4.42	26.73±0.75	2.994±0.002	3.150±0.006	3.407±0.002	3.005±0.003
L44	39.32±2.25	23.21±0.84	2.962±0.003	3.142±0.005	4.617±0.001	4.055±0.014
L45	40.81±3.44	18.15±0.91	4.581±0.005	5.369±0.018	4.562±0.001	4.107±0.027
L46	39.42±4.82	36.09±2.04	2.334±0.003	2.735±0.010	4.539±0.001	3.060±0.013
L47	41.70±0.51	31.92±1.04	2.987±0.005	3.122±0.004	4.741±0.005	3.905±0.002
L48	44.12±3.06	39.99±4.18	3.612±0.004	4.408±0.006	4.646±0.006	4.014±0.003
L49	41.58±4.26	20.66±0.79	3.704±0.003	4.556±0.011	4.614±0.003	3.020±0.003
L50	44.26±4.11	39.49±2.11	2.324±0.004	2.937±0.006	3.963±0.008	2.068±0.007
L51	29.96±1.01	28.92±0.87	2.356±0.003	2.466±0.007	4.670±0.001	3.539±0.022
L52	42.72±0.95	30.13±1.07	2.546±0.002	2.701±0.006	4.516±0.001	3.855±0.024
L53	40.95±3.59	36.35±2.46	2.910±0.003	4.764±0.021	4.443±0.005	2.543±0.004
L54	49.68±1.00	40.43±0.76	2.790±0.002	9.927±0.003	4.290±0.004	4.656±0.008
L55	43.03±4.54	28.23±1.94	4.157±0.006	4.685±0.011	4.063±0.006	3.744±0.026
L56	40.26±3.19	36.23±4.18	5.224±0.013	6.810±0.041	4.654±0.003	3.632±0.011
L57	41.17±5.80	30.45±1.97	4.841±0.003	4.822±0.013	4.200±0.002	3.053±0.002
L58	40.87±1.12	17.39±1.34	3.160±0.003	3.562±0.002	4.521±0.005	4.135±0.004
L59	31.15±2.72	30.21±1.17	3.644±0.003	3.725±0.004	4.567±0.002	3.415±0.003
L60	40.54±3.30	37.80±4.56	2.779±0.004	2.997±0.006	4.402±0.001	4.023±0.005
L61	35.52±0.97	26.57±4.66	4.232±0.005	5.541±0.004	4.733±0.006	4.156±0.010
L62	36.43±0.94	17.70±1.37	3.049±0.002	3.149±0.004	4.590±0.004	3.721±0.004
L63	43.28±2.36	30.96±2.13	5.613±0.023	7.469±0.001	4.734±0.008	4.418±0.008
L64	37.07±3.26	28.52±2.39	4.016±0.003	3.815±0.004	4.546±0.005	3.550±0.013
L65	42.54±1.63	31.67±0.89	3.022±0.003	3.910±0.007	4.103±0.001	4.762±0.003
L66	46.72±3.01	32.48±2.13	4.016±0.003	4.929±0.004	4.567±0.005	4.316±0.001
L67	35.66±3.12	19.39±0.45	2.911±0.108	3.148±0.002	4.414±0.001	3.172±0.004
L68	34.95±4.31	22.84±1.17	3.158±0.002	4.013±0.005	4.121±0.001	3.481±0.001
L69	32.55±1.53	24.08±4.10	2.457±0.006	2.739±0.005	3.470±0.001	2.931±0.005
L70	42.50±2.04	23.56±2.21	2.747±0.002	2.826±0.005	4.434±0.001	4.260±0.004
L71	36.60±3.85	25.51±2.53	3.902±0.012	4.223±0.002	4.487±0.001	3.481±0.003
L72	49.45±2.74	32.36±2.33	3.560±0.007	3.753±0.004	4.628±0.001	4.112±0.006
L73	36.72±3.81	27.98±1.28	3.353±0.004	3.350±0.008	4.452±0.001	2.481±0.002
L74	29.29±2.27	20.76±3.39	3.412±0.003	4.529±0.002	4.380±0.004	2.571±0.003
L75	30.21±5.12	23.11±1.16	2.548±0.003	2.745±0.001	4.571±0.001	4.559±0.003
L76	43.41±2.56	27.33±1.05	3.060±0.004	3.129±0.006	4.354±0.001	3.598±0.027
L77	52.45±5.59	24.07±2.27	3.621±0.002	3.657±0.007	4.583±0.001	3.876±0.014
L78	40.05±4.95	23.37±2.40	2.941±0.003	3.111±0.014	4.592±0.001	2.581±0.039
L79	23.94±3.10	10.35±0.61	3.225±0.010	3.664±0.001	4.389±0.001	3.601±0.027
L80	43.72±1.97	32.93±2.02	3.008±0.004	3.130±0.014	4.062±0.000	2.887±0.026
L81	27.41±1.36	25.41±1.39	2.468±0.004	2.519±0.015	4.730±0.001	4.683±0.004
L82	29.63±1.48	19.43±0.57	3.497±0.004	3.476±0.068	3.763±0.001	3.721±0.037
L83	49.49±3.99	22.24±1.20	3.4618±0.002	3.539±0.009	4.436±0.001	3.856±0.012

Supplementary Table S3. Effect of moisture deficit stress on proline content (µg/gm) and protein content (µg/ml) in melon parents and population under well water (WW) and moisture deficit stress (MS) conditions

Genotype	WW	Proline MS	Per cent increase	WW	Protein MS	Per cent decrease
P1	4.01±0.00	4.82±0.02	20.13	4.62±0.00	4.11±0.03	10.99
P2	2.54±0.00	8.67±0.00	240.79	4.68±0.00	4.30±0.42	8.00
F_1	2.75±0.00	3.04±0.00	10.53	4.54±0.00	4.05±0.01	10.79
$F_{2:3}$	3.09± 0.37	3.77±0.72	22.10	4.39±0.22	3.76±0.44	14.43

The values are mean of three replicates ± standard error

Supplementary Table S4. Analysis of Variance for chlorophyll content and leaf area traits in melon genotypes

		Chlorophyll content			Leaf area		
Source of var.	**DF**	**SS**	**MS**	**F-value**	**SS**	**MS**	**F-value**
Replication	2	390.29	195.14	15.42	64.54	32.27 ^{ns}	1.40 ^{ns}
Genotype	85	3650.95	42.95**	3.39**	23058.3	271.2**	11.83**
Drought	1	49.42	49.42	3.90	5894.50	5894**	257.0**
G × D	85	3053.87	35.92**	2.84**	13769.2	161.9**	7.06**
Trt SS	171	6754.25	39.49**	3.12**	42722.1	249.8**	10.89**
Error	342	4326.43	12.65		7841.30	22.92	
Total	515	11470.98			50627.9		

ns = non significant, SS = sum of square, MS = mean square
DF = degree of freedom, *significant at ($p \leq 0.05$) , **significant at ($p \leq 0.01$)

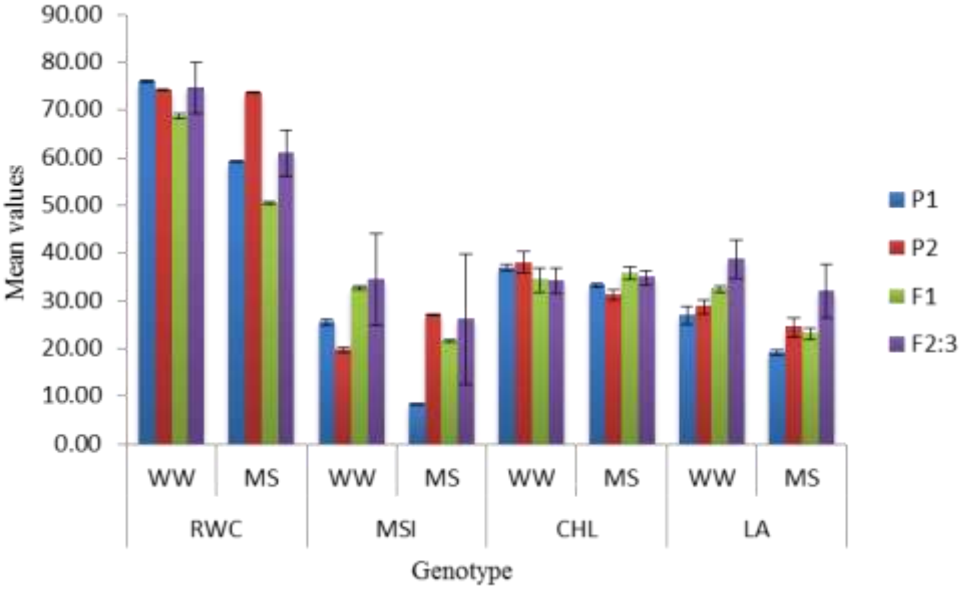

Fig. 1. Variation in mean value of three replicates ± standard error for relative water content (RWC) (%), membrane stability index (MSI) (mS/cm), chlorophyll (Chl) (SPAD unit) and leaf area (LA) (cm^2) of melon parents and population under well water (WW) and moisture deficit stress (MS) conditions