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RESEARCH ARTICLE

Evaluation of salinity response through the antioxidant defense system and osmolyte accumulation in a mutant rice

Maryam Forough[1], Saeid Navbpour [*1], Esmaeil Ebrahimie[2], Ali Akbar Ebadi[3] and Davood Kiani[4]

[1]Plant Breeding and Biotechnology Dept. Gorgan University of Agricultural Sciences and Natural Resources. Gorgan, Iran
[2]Genomics Research Platform, School of Life Sciences, La Trobe University, Melbourne, Victoria 3086, Australia
[3]Rice Research Institute of Iran (RRII), Agricultural Research Education and Extension Organization (AREEO) Rasht, Iran
[4]Seed and Plant Improvement Research Department, Bushehr Agricultural and Natural Resources Research and Education Center, AREEO, Bushehr, Iran

ABSTRACT: In order to assess the responses of Hashemi rice genotype and its advanced mutant line under salinity stress of 100 mM Sodium chloride (NaCl) for three and six days the shoot samples were taken for biochemical analysis. This experiment was performed in split plot based on randomized complete block design with three replications. The main factor was factorial combination of saline treatment and sampling period, sub factor included genotypes. The result showed that the chlorophyll content decreased (16.3) under salt stress for the wild type, but higher amount (21.2) in the mutant was recorded. The mutant rice showed higher amount of K^+ and lower of Na^+ concentrations in shoots under salt stress condition. The results revealed, although the amount of H_2O_2 of both genotypes was significantly increased by exposure to NaCl, the effect was superior in the wild genotype (44.85). The antioxidant enzymes activity include catalase and peroxidase activity were grow up significantly in advanced mutant line. Also, the level of flavonoids and phenol content under salinity stress were enhanced dramatically in mutant line. In order to evaluate ion homeostasis under salinity stress condition the measurement of osmolytes such as proline, glycine betaine and trehalose indicated the mutant rice by rising the production (4.4, 0.81 and 87.55 respectively) of these metabolites in shoot showed the better tolerance to salinity stress. In conclusion, the observation indicated that mutation had a positive impact on ROS scavenging system and ion homeostasis mechanism and ultimately have led to salt tolerance in the mutant genotype.

KEYWORDS: Enzyme activity, Ion homeostasis, Mutation, Sodium chloride

INTRODUCTION

Rice (*Oryza sativa* L.) is known as a critical food source of near one-half of the world's population but salinity is a great threat for the rice production [66]. Salinity of soil or water mainly due to high concentrations of Sodium chloride (NaCl) considered as one of the most deleterious abiotic stress that threat crop growth and productivity significantly [27] and rice is a sensitive to this stress particularly at the early vegetative state [21]. Moreover, salt tolerance is a complex trait and tolerant plants could survive in facing of this stress by altering and developing

*Corresponding author (✉): s.navabpour@yahoo.com
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in some physiological and biochemical process [46]. One of the crucial consequences of exposure to salinity stress in plants, is to disturb the cellular homeostasis and increase the ROS such as hydrogen peroxide (H_2O_2) superoxide anion (O_2^-) and hydroxyl radicals ($OH^·$) and ultimately leads to oxidative stress [45]. The higher intracellular concentration of various ROS rather than rate of their scavenging can oxidize DNA, protein, and lipids and impair enzymatic activities due this they are supposed to be toxic to plant cells [13]. In order to avoid of this harmful consequence of accumulation ROS, plant cells developed array of mechanisms involving enzymatic and non-enzymatic processes which showed magnificent protective role in tolerance to extreme environments particularly salinity stress [41, 22]. In enzymatic manner, plants could regulate the amount of ROS through the activity of glutathione reductase, catalase, ascorbate peroxidase, superoxide dismutase and glutathione peroxidase. Superoxide dismutase is a key scavenger of O_2^-, and its enzymatic function consequence in the production of H_2O_2 and O_2. At this point, the hydrogen peroxide generated is scavenged by catalase and some other peroxidases. Catalase, convert H_2O_2 into water and molecular oxygen [17]. The enzyme glutamate dehydrogenase represents important role in amination and deamination reactions in the plant tissues [35]. Non-enzymatic system comprises antioxidants such as phenol and flavonoids to scavenge ROS [49].

Beside the oxidative stress, the higher concentration of NaCl stimulates osmotic stresses that diminishes the capability of plants to absorb water and minerals [29]. Under this condition plant synthesize and accumulate osmolytes in the cytoplasm and in chloroplasts which is critical to maintain cellular water and cope stress condition. The osmolytes compounds are highly soluble and have low molecular weight such as proline and glycine betaine. The Osmolytes perform multiple functions during stress particularly as osmotic adjustment and scavenging of reactive oxygen species (ROS), membrane integrity maintains, and stabilization of enzymes and proteins [64, 12, 8]. Moreover, increase the Na^+ and $Cl^–$ concentrations which move within plants by transpiration stream and accumulate in leaf tissue is another noticeable salinity stress consequence and ROS signaling has a key role in regulating and controlling shoot Na^+ accumulation through regulating vasculature Na^+ concentrations [28]. This phenomenon could have a lethal effect on many cellular processes particularly protein–protein interactions and electrochemical gradients. It has been exhibited the rate of Na^+/K^+ efficiently improve the salinity tolerance [11, 15].

Generally, study on the response to salinity stress would be helpful in breeding plant salt tolerant cultivars by identifying physiological features. The goal of the present study is to provide evidences that gamma radiation has influenced on antioxidant defense system and osmolyte accumulation and caused to improve salinity tolerant over seedling stage rather than wild type. After field assessment and finding tolerant mutant lines it will be so useful through biochemical and physiological analysis try to better evaluate to salinity tolerance. The results will be revealed how mutation has influence and improved salinity tolerant trait. Ultimately, we have tried to evaluated salinity tolerance based on the rate of production and activity of antioxidant enzymes in detoxification of reactive oxygen species and synthesis of compatible solutes.

MATERIALS AND METHODS

Plant materials and Salt treatment

This experiment was carried out at Gorgan University of Agricultural Sciences and Natural Resources. The two genotypes of rice, the wild-type genotype “Hashemi” and its advance mutant line, were provided from Rice Research Institute, Rasht, Iran. The mutant genotype was generated by gamma irradiation and assessed for salt tolerance in farm trials with EC 8 dS m^{-1}. Healthy and uniform seeds of both the genotypes were chosen, and their surfaces were sterilized using 2% sodium hypochlorite and were put in petri dishes with 10 ml water and filter paper at 23 ±1 ℃ for six days under dark condition. On top of this the germinated seeds were transferred in to the specific container of Youshida solution [65]. In order to adjust the pH of solution to 5.8 NaOH was used. The Seedlings grew up in controlled condition with 22-26 ℃ 16 h day and 8 h night and 65% humidity. Three biological replicates were used for control and 100 mM NaCl (Merck, Germany) treatment. After six days grow under the mentioned normal condition, 100 mM NaCl was added as a salinity stress and throughout two interval times three and six days after salinity stress samples from shoots were collected from both normal and salinity conditions. The samples were immediately frozen in liquid nitrogen and kept at -80 ℃ for further biochemical analysis.

Ion measurements

In order to detect the amount of sodium and potassium, the Flame photometer method was done [63].

Measurement of H_2O_2 and lipid peroxidation

For the quantification of hydrogen peroxide and lipid peroxidation levels, tissue of shoot (0.5 g) were homogenized in 5% trichloroacetic acid (TCA) and then used for measurement of hydrogen peroxide (H_2O_2) amount through the manner of Sagisaka (1976). The amount of lipid peroxidation was detected with 2-thiobarbituric acid (TBA) reactive metabolites chiefly malondialdehyde (MDA) accumulation [24].

Enzyme Extraction and Assays

The amount of superoxide dismutase (SOD) (EC 1.15.1.1) enzymatic activity was detected according to Beyer *et al.* [10]. Catalase activity (CAT; EC 1.11.1.6) was measured by Aebi and Lester's method [4] based on the consumption of H_2O_2. Ascorbate Peroxidase (APX; EC 1.11.1.11) activity was examined using a modified manner of Nakano and Asada [48]. APX activity was determined from the reduction in absorbance at 290 nm as a result of oxidation of ascorbate in the reaction. Glutathione reductase expression (GR; EC 1.8.1.7) was analyzed as reported by Smith *et al.* [58] method and lipoxygenases activity (LOX; EC 1.13.11.12) was measured in consonance with Ikuo's modified method [25].

Measurement of non-oxidant enzymes and chlorophyll

The level of chlorophyll was measured by Lichtenthaler's [39] method. To measure the total phenolic, the method of Soland *et al.* [59] was performed. The Flavonoid contents were measured through Zhishen *et al.* [67].

Determination of osmolytes

Proline content was elucidated by Gilmour *et al.*, [23]. The content of trehalose in rice shoot were determined by following the method described by Li *et al.*, [2014] with some modifications. The glycine betaine was assayed according to previously described methods (Hitz and Hanson 1980). Total soluble sugar was assayed based on Dubosi et al [19] method.

Statistical Analysis

This experiment was conducted in split plot based on randomized complete block design with three replications. The main factor was factorial combination of saline treatment and sampling period, sub factor included genotypes. All biochemical measurements data were subjected to analysis of variance (ANOVA) and the means were compared by LSD test at the 5% level using SAS 9.1 software. In order to show differences between genotypes under salinity and normal stress, all bar charts were depicted by T-test and Excel 2017 software.

RESULTS and DISCUSSION

The most soluble salt that was finding in soil to cause salinity is NaCl. Plants under salinity stress raise tissue Na^+ accumulation and K^+ loss [28, 69,46]. Restriction of Na^+ accumulation and not decreasing K^+ amount is well known as a salinity tolerance mechanism [28]. Significant changes in Na^+, K^+ concentration and Na^+/K^+ ratio (Table 1) in shoot of the two genotypes in saline culture were observed. Sodium accumulated to a much greater extent in wild genotype than in the mutant rice and in other hand significant higher accumulation K^+ in mutant rice was determined. Furthermore, Na^+/K^+ ratio was higher in the shoot of mutant rice which strongly indicated the mutant rice showed better salinity tolerance. Exceptional accumulation of sodium leads to numerous toxic effects like distraction of membrane stability, lipid peroxidation and total chlorophyll concentration and photosynthetic capacity as well as yield components. In many researches were indicated that the salt tolerant genotypes have higher Na^+ exclusion and K^+ accumulation [54, 30].

Many studies have been demonstrated the increase of hydrogen peroxide (H_2O_2) over exposure to a salinity stress, and the intensity and duration of the stress are key factors in its production [57]. The result of this study, presented that aggregation of H_2O_2 in the shoot of wild rice was more extra (1.27-fold) than the mutant rice after six days of salinity (Fig. 1).

In our study substantial declines in chlorophyll content under salt stress in comparison with control condition was observed although higher amount belongs to mutant genotype significantly (Fig. 2). Drop in chlorophyll contents mainly because of reducing light absorbance as a photoprotection mechanism over salinity stress [20]. Previous studied indicated that chlorophyll content in plants affect directly on the plant health and growth and

Table 1: The mean comparison of both genotypes under salinity stress for ions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

Traits	Genotype	3 days		6 days	
		Normal	salinity	Normal	salinity
Na^+	W	8.2±0.4 a (d)	224±1.7 a (b)	12±0.5 a (d)	237±1.1 a (a)
	M	7.25±0.4 a (d)	207.5±2.0 b (c)	11±0.5 a (d)	242±4.0 a (a)
K^+	W	405±2.8 b (d)	282±1.7 b (f)	414±0.5 b (c)	269±1.1 b (e)
	M	441.5±2.0 a (b)	306±2.8 a (e)	452±1.7 a (a)	287.5±4.3 a (e)
Na^+/K^+	W	8.2±0.4 a (e)	224±1.7 a (c)	12±0.5 a (e)	237±1.1 a (a)
	M	7.25±0.4 a (e)	207.5±2.0 b (d)	11±0.5 a (e)	242±4.0 b (a)

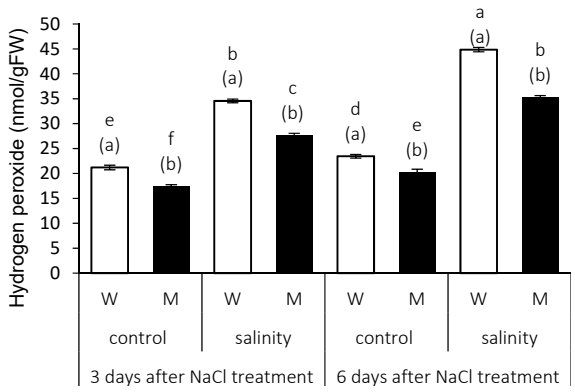

Fig. 1. The mean comparison between the rice genotypes for the hydrogen peroxide level under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

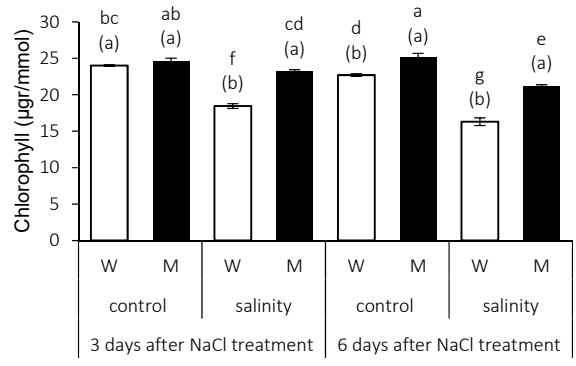

Fig. 2. The mean comparison between the rice genotypes for the chlorophyll content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

total chlorophyll content decreases in rice [6] and *Vigna subterranean* L. [60] under NaCl salinity.

As one of the harmful consequences of over production of hydrogen peroxide under salinity stress was lipid peroxidation. The amount of malondialdehyde produced by lipid peroxidation of cell membrane indicated the salt and oxidative harms and damages [42]. The malondialdehyde concentrations of salt-stressed seedlings grew up in comparison with the control condition (Fig. 3). The amount of lipid peroxidation was 1.12 and 1.23 times higher in wild type in compare with mutant in first and second sampling time respectively in saline condition and these observations indicated the mutant rice has more ability to remove ROS. Furthermore, the amount of lipoxygenase enzyme in mutant rice was significantly greater (3.07-fold) than wild during after 6 days of salinity stress which could contribute in lipid peroxidation of membrane lipids and reduce it significantly (Fig. 4). By this fact lipoxygenase activity has a crucial role for the oxidation of polyunsaturated fatty acids and therefore improves lipid peroxidation under stress conditions [50]. Chutipaijit *et al.* [16] also observed that in the resistant to salinity of rice seedling the amount of malondialdehyde was significantly less than those sensitive and their results indicated that seedlings with less malondialdehyde had better efficiency to endure the damage of cellular membranes under salinity. In other different studies showed salt tolerant Sesame cultivar [31] and salt tolerance *Phaseolus vulgaris* L [61] as well as halophyte *Cakile maritima* [33] had lower levels of lipid peroxidation which could be essential mark of reduction oxidative damage over salinity. The excess of ROS is destructive to the plant life due this fact to make equilibrium between ROS production and scavenging is crucial for redox homeostasis [7]. Plants with various antioxidant enzymes try to regulate the ROS levels [46].

Table 2: The mean comparison of both the genotypes under the normal and salinity stress conditions for antioxidant enzymes (SOD: Superoxide dismutase, CAT: Catalase, APX: Ascorbate peroxidase, GR: Glutathione reductase and GPX: Glutathione peroxidase). The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between parent and mutant genotypes under stress and sampling conditions (LSD = 0.05).

Traits	Genotype	3 days		6 days	
		Normal	salinity	Normal	salinity
SOD	W	20.2±0.4 a (g)	31.45±0.3 b (c)	21.65±0.3 b (f)	27.8±0.3 b (d)
	M	21±0.3 a (fg)	41.2±0.6 a (a)	23.6±0.2 a (e)	35.75±0.3 a (b)
CAT	W	39.9±0.0 b (e)	36.55±0.4 b (f)	40.5±0.2 b (e)	25.8±0.3 b (g)
	M	44.7±0.1 a(d)	59.8±0.4 a (b)	47.2±0.5 a (c)	65.45±0.7 a (a)
APX	W	0.46±0.0 b (e)	0.74±0.0 b (c)	0.49±0.0 b (e)	0.76±0.0 b (c)
	M	0.58±0.0 a (d)	0.97±0.0 a (b)	0.62±0.0 a (d)	1.08±0.0 a (a)
GR	W	32.55±0.3 b (e)	37±0.9 b (cd)	33.1±0.5 b (e)	21.55±0.8 b (f)
	M	36.65±0.7 a (d)	57.1±0.4 a (b)	39.1±0.5 a (c)	63.35±1.1 a (a)
GPX	W	36±0.5 b (g)	52.3±0.6 b (b)	34.8±0.1 b (g)	50.2±0.4 b (b)
	M	40.45±0.6 a (f)	66.8±0.4 a (b)	42.3±0.1 a (e)	73±0.2 a (a)

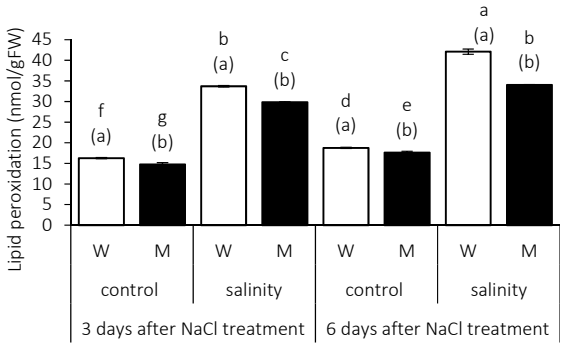

Fig. 3. The mean comparison between the rice genotypes for the lipid peroxidation under the normal and salinity stress conditions. The means with the same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

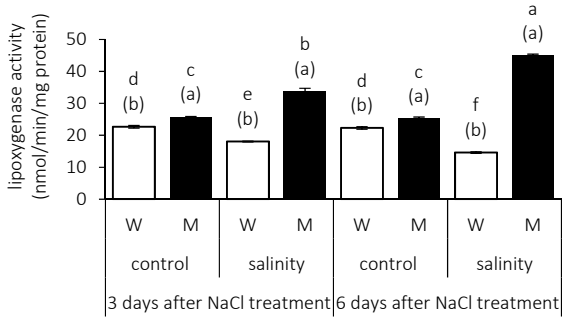

Fig. 4. The mean comparison between the rice genotypes for the lipoxygenase activity under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

Through ROS decontamination procedure, it was observed that superoxide dismutase is as the first enzymes among antioxidant enzymes which catalyzing the superoxide anions detoxification [44]. The observations of this research showed the higher expression of superoxide dismutase in mutant rice (Table 2). superoxide dismutase act in early of stress period and use an oxygen free radical as its substrate (superoxide) and then catalyzes the disproportionation of superoxide radicals (O_2^-) into molecular oxygen and hydrogen peroxide (43, 53]. It has been studied in different plants such as Arabidopsis [55] wheat [62] which are more tolerant to salinity stress, the superoxide dismutase activity has grown up in them.

The hydrogen peroxide (H_2O_2) produced by superoxide dismutase, can be scavenged by catalase which is found in the plants and other aerobic organisms. In fact, catalase convert hydrogen peroxide into harmless product includes water and oxygen [56]. The mutant rice indicated more activity of catalase significantly in compare with wild genotype under salinity stress (Table 2). Higher expression of catalase under salinity in different plant species such as *Nerium oleander* [34] and *Cucumis sativus* [68] has been determined.

Beside the catalase, plants have another set of H_2O_2 scavenging enzymes such as ascorbate peroxidases and glutathione peroxidases which are distributed and found in many cellular compartments [45]. The activity of ascorbate peroxidases and glutathione peroxidases under salinity stress has been investigated in our study and observation demonstrated of higher activity of these enzymes in mutant genotype (Table 2). The content of

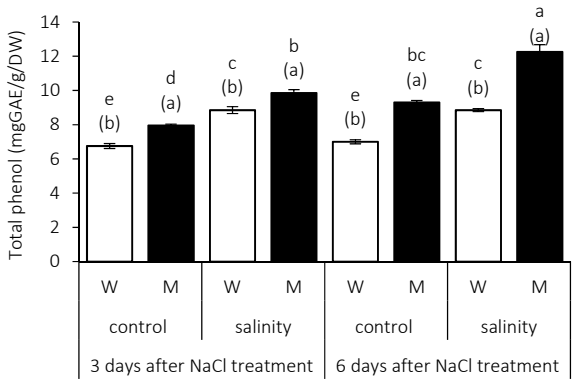

Fig. 5. The mean comparison between the rice genotypes of the total phenolic content under the normal and the salinity stress conditions. The means with the same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

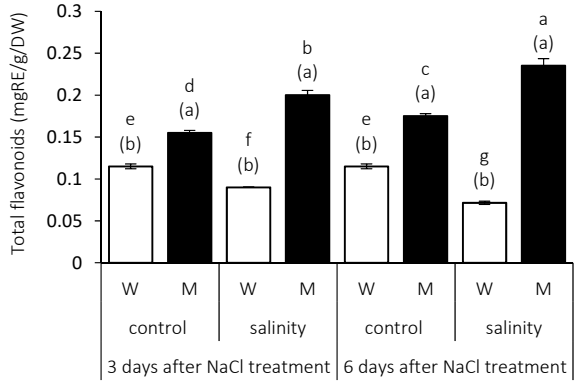

Fig. 6. The mean comparison between the rice genotypes for the total flavonoid content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

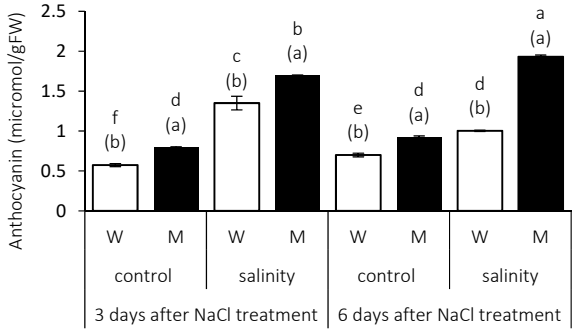

Fig. 7. The mean comparison between the rice genotypes for the anthocyanin content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

ascorbate peroxidase and glutathione peroxidase in mutant rice on sixth day of salinity (1.42-fold and 1.45-fold upper respectively) than wild genotype. It has been demonstrated there is a strong correlation between glutathione peroxidase activity and detoxifying lipid hydroperoxides and other reactive molecules in different species and under several stress conditions particularly salinity [18]. Similarly, for glutathione reductase activity mutant rice showed higher (2.93-fold) than wild (Table 2).

In addition of antioxidant enzyme activity, measurements of non- antioxidant enzyme such as total flavonoid, total phenol and anthocyanin were performed. The consequence of salt stress on the accumulation of total phenolic compounds in the shoot of mutant seedlings which were higher than wild type (Fig. 5). It has been demonstrated that the accumulation of phenolic compounds causes inhibition of lipid peroxidation [52]. Likewise, rise in phenolic compounds in response to salinity has also been observed in extract of various plants [14, 40, 2]. These studies indicated that the phenolic compounds protect plants against the oxidative stress created by salinity and leads to enhance the salinity tolerance.

In this investigation under the salinity stress treatment the amount of total flavonoid surged as well (Fig. 6). Previously it has been proved that since flavonoids have hydroxyl groups in their structure, it could scavenge the reactive oxygen species (ROS) and inhibit the ROS production [5, 26]. Also, flavonoids have key role in maintain membrane integrity by binding to the polar head groups of phospholipids and accumulate at the membrane surfaces [52]. Hence, the findings suggest that higher flavonoids in the mutant genotype could show more protective role against salinity stress.

Anthocyanin has a higher level of hydroxylation due this it has the superior anti-oxidative capability [47]. In this present study, the accumulation of anthocyanin grows up under salinity stress which was attributed to enhance salinity tolerance in mutant genotype (Fig. 7). This result, along with Bandurska *et al.,* [9] clearly showed correlation of anthocyanin with salinity tolerance.

When the salt concentration raises up, water potential reduces and, plants try to enhance the osmotic potential consequently. So, osmotic stress is one of the outputs of salinity stress. In this situation plants by accumulations of high concentrations of inorganic ions or low molecular weight organic solutes able to cope the osmotic stress [21].

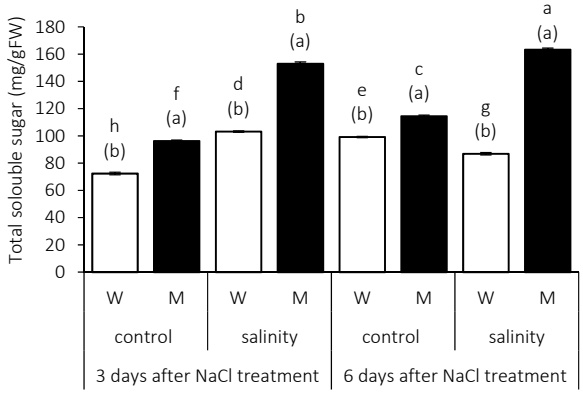

Fig. 8. The mean comparison between the rice genotypes for the soluble sugars content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

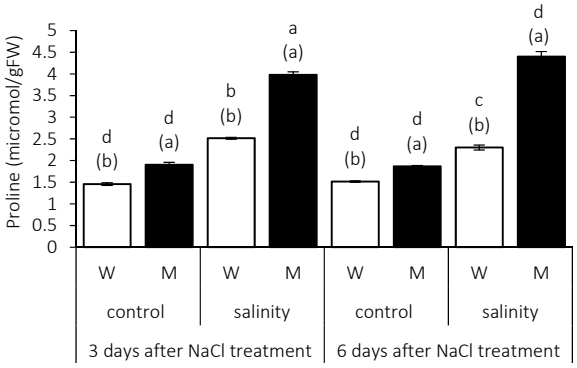

Fig. 9. The mean comparison between the rice genotypes for the proline content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

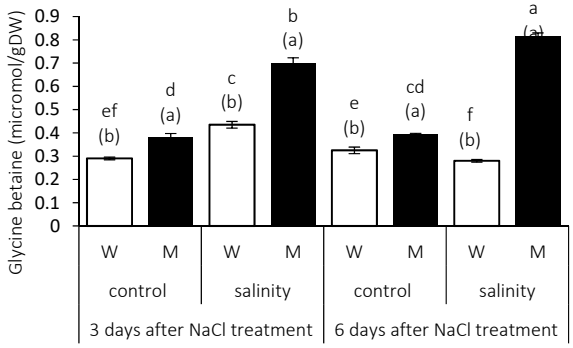

Fig. 10. The mean comparison between the rice genotypes for the glycine betaine content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

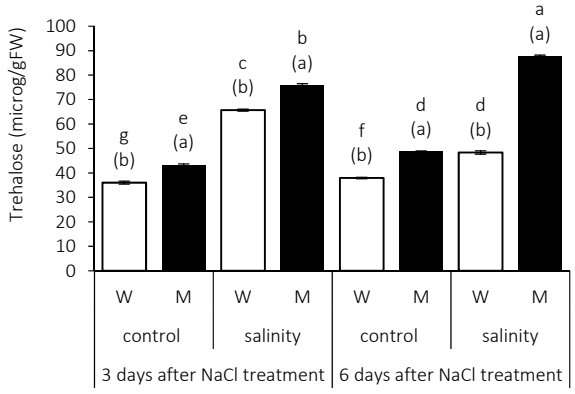

Fig. 11. The mean comparison between the rice genotypes for the trehalose content under the normal and salinity stress conditions. The means with same letters had no significant differences statistically (LSD = 0.05). The same letters in parentheses indicate no statistically significant difference between the parent and mutant genotypes under the stress and sampling conditions (LSD = 0.05).

In this study some of the different osmolytes were assayed in response to salinity. The accumulation of soluble sugars in the mutant rice was significantly greater than wild type (Fig. 8) and this had a huge impact on the improvement osmoprotection and also source of energy for creating range of cell compartments and metabolites [52]. An extremely significant influence of salt on the increase of proline and glycine betaine in rice seedlings were detected in mutant rice. The proline accumulation was detected for shoot mutant rice (1.91-fold) notably higher than wildtype under 100 mM NaCl (Fig. 9). In the same condition for the glycine betaine content in mutant rice were higher (2.89-fold) than wild type (Fig. 10). This high amount of proline glycine betaine may reflect of greater osmoregulation in mutant genotype and leads to more tolerant to salinity stress [3].

Trehalose, a natural non-reducing sugar, also is another compatible osmoprotectant and the results of this research revealed that the mutant genotype had a higher trehalose content in shoot the highest amount of trehalose was 1.8 fold greater than wild type (Fig. 11) and it has been indicated that accumulation of trehalose associated with reduction of oxidative damage by dropping the rate of ROS production [32]. Abdallah *et al.*, [1] showed salinity on seedlings which were treated with 25 mM trehalose on pre sowing seed, leads to decrease the photosynthetic pigments, proline, trehalose and total soluble sugar contents however higher activity of antioxidant enzymes were recorded. Authors indicated that seed priming with trehalose is effective to protect rice in salinity stress.

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ارزیابی پاسخ به شوری با بررسی سیستم دفاعی آنتی اکسیدانی و تجمع اسمولیت‌ها در برنج جهش یافته

مریم فروغ[1]، سعید نواب پور[2]*، اسماعیل ابراهیمی[3]، علی اکبرعبادی[4]، داوود کیانی[5]

[1]گروه اصلاح نباتات و بیوتکنولوژی، دانشگاه علوم کشاورزی و منابع طبیعی گرگان

[2] گروه اصلاح نباتات و بیوتکنولوژی، دانشگاه علوم کشاورزی و منابع طبیعی گرگان

[3]مرکز تحقیقات ژنومیک، دانشکده علوم زیستی، دانشگاه لاتروب، ملبورن، ویکتوریا ۳۰۸۶، استرالیا

[4]موسسه تحقیقات برنج کشور، سازمان تحقیقات آموزش و ترویج کشاورزی، رشت، ایران

[5]بخش تحقیقات اصلاح و تهیه نهال و بذر ، مرکز تحقیقات و آموزش کشاورزی و منابع طبیعی استان بوشهر، سازمان تحقیقات، آموزش و ترویج کشاورزی، بوشهر، ایران

نویسنده مسئول: s.navabpour@gau.ac.ir

چکیده

به منظور ارزیابی پاسخ ژنوتیپ برنج هاشمی ولاین جهش یافته پیشرفته آن تحت تنش شوری ۱۰۰ میلی متر کلرید سدیم (NaCl) به مدت سه و شش روز از نمونه های ساقه‌چه برای تجزیه و تحلیل بیوشیمیایی استفاده شد. این آزمایش در قالب کرت اسپلیت بر اساس طرح بلوک‌های کامل تصادفی با سه تکرار انجام شد. فاکتور اصلی ترکیب فاکتوریل از تیمار شوری و زمان نمونه‌برداری و فاکتور فرعی شامل ژنوتیپ‌ها بود. نتایج نشان داد که میزان کلروفیل تحت تنش شوری برای ژنوتیپ والد (۱۶/۳) کاهش یافته است، اما به میزان بیشتر (۲۱,۲) درژنوتیپ جهش یافته گزارش شد. برنج جهش یافته غلظت بیشتری از K^+ و غلظت کمتری از سدیم را درساقه‌چه‌های تحت شرایط تنش شوری نشان داد. نتایج نشان داد، اگرچه میزان H_2O_2 هر دو ژنوتیپ با قرار گرفتن در معرض NaCl به طور معنی داری افزایش یافت ، اما مقدار آن در ژنوتیپ وحشی بیشتر بود (۴۴/۸۵). فعالیت آنزیمهای آنتی اکسیدانی شامل کاتالاز و فعالیت پراکسیداز به طور قابل توجهی در لاین جهش یافته پیشرفته افزایش یافت. همچنین، مقدار فلاونوئیدها و فنل تحت تنش شوری در لاین جهش به طور چشمگیری افزایش یافت. به منظور ارزیابی هموستاز یونی در شرایط تنش شوری، اندازه گیری اسمولیت ها از جمله پرولین ، گلیسین بتائین و ترهالوز نشان داد که برنج جهش یافته با افزایش تولید این متابولیت‌ها (به ترتیب ۴/۴ ، ۰/۸۱ و ۸۷/۵۵) در ساقه‌چه‌ها تحمل بهتری نسبت به تنش شوری نشان داد. در نتیجه، این مشاهدات حاکی از آن است که جهش تأثیر مثبتی بر سیستم اصلاح گونه‌های فعال اکسیژن و مکانیسم هموستاز یونی داشته و در نهایت منجر به تحمل به شوری در ژنوتیپ جهش یافته شده است.

کلمات کلیدی: فعالیت آنزیمی، سدیم کلرید، موتاسیون، هموستازی یونی