Journal of

Plant Sciences

ISSN 1816-4951

Academic Journals Inc.

www.academicjournals.com

Journal of Plant Sciences 2 (2): 141-152, 2007
ISSN 1816-4951

Exogenous Glucose and Abscisic Acid Pre-treatment in Indica Rice (*Oryza sativa* L. spp. *indica*) Responses to Sodium Chloride Salt Stress

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Abstract: The aim of this research is to investigate on the osmotic adjustment and pigment preservation by soluble sugar accumulation in salt stressed rice using exogenous glucose and abscisic acid (ABA) application, leading to plant growth and development. Soluble sugars including sucrose, glucose and fructose in the salt-stressed root tissues were continuously increased in the conditions of 111-222 mM glucose and 20-60 µM ABA treatments and then drop in the extreme 333-444 mM glucose and 80 µM ABA treatments. Osmolarity in the salt-stressed root tissues showed the similar pattern to the sugar responses and was negatively related to soluble sugar concentration (r = 0.91). Chlorophyll a, chlorophyll b and total carotenoid concentrations in the salt-stressed seedlings were significantly maintained by endogenous sugar osmotic adjustment. In addition to, the high osmolarity in salt-stressed seedlings was negatively related to total chlorophyll stabilization (r = 0.83). The total chlorophyll degradation in the salt-stressed leaf tissues was positively correlated with plant growth defined by shoot height (r = 0.81). Root length, root number and root cortex thickness of salt-stressed rice seedlings showed the highest at 222 mM glucose and 60 µM ABA treatments for 146.1 cm, 17.3 and 1.3 µm, respectively. An exogenous application of glucose and ABA in this investigation is an alternative way to acclimatize the rice crop before exposed to soil salinity and further applied for rice cultivation in salinity filed trial.

Key words: Chlorophyll degradation, cortex thickness, osmolarity, soluble sugar, salinity

INTRODUCTION

Rice is an important carbohydrate crop to supply more than half of world's population, especially in Asia. Rice crop is cultivated and consumed in Asian countries for 90%, which is a major food crop (Khush, 2005). Main barrier of rice production is abiotic stresses such as salinity soil, water deficit, flooding, nutrient deficiency/enrichment, acidic/basic soils, extreme temperature and UV irradiation (Cushman and Bohnert, 2000; Salekdeh *et al*., 2002; Lafitte *et al*., 2004). In case of soil salinity, there are several regions about 45.4 million hectares, including Asia, Africa, Latin America, North America, Europe and Australia, to be a serious problem, especially irrigated areas (Ghassemi *et al*., 1995; Singh and Chatrath, 2001). Rice crop has been reported as salt-sensitive species, especially in seedling and flowering stages, leading to yield reduction (Shannon *et al*., 1998; Zeng and Shannon, 2000; Khan and Abdullah, 2003; Zeng *et al*., 2002). Salt-tolerant breeding program in rice is a fruitful topic for plant breeders to solve the salinity toxic damage in both osmotic and ionic

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effects(Gregorio *et al*., 2002; Senadhira *et al*., 2002; Flowers and Flowers, 2005). It should be used an ntensive skill, time requirement and wide genetic resources. Alternatively, an exogenous application of osmotic solutes i.e., glycinebetaine (Cha-um *et al*., 2006; Demiral and Turkan, 2006), putrescine, spermine (Ndayiragije and Lutts, 2006) and spermidine (Roy *et al*., 2005; Ndayiragije and Lutts, 2006) has been reported. In addition to, ABA and sugar exogenous applications have been pretreated or acclimatized the plant before exposed to extreme environmental conditions, including salinity and drought (Gibson, 2000; Lin and Kao, 2001; Wang *et al*., 2003; Yin *et al*., 2004; Morsy *et al*., 2006). However, the function roles of exogenous sugar and ABA in salt defense responses are still unclear. Pathumthani 1 rice is a salt sensitive variety of indica subspecies, which is photoperiod insensitive, aroma flavor, high cooking quality and high yield (Ariyaphanphitak *et al*., 2005; Leohakunjit and Kerdchoechuen, 2007). In present study, the salt-tolerant defense mechanisms in *indica* subspecies using *in vitro* glucose and ABA application were intensively evaluated.

MATERIALS AND METHODS

Plant Material

Seeds of *indica* rice (*Oryza sativa* L. spp. *indica* cv. Pathumthani 1) were obtained from the Pathumthani Rice Research Center (Rice Research Institute, Department of Agriculture, Ministry of Agriculture and Cooperative, Thailand). Seeds were dehusked by hand, sterilized once in 5% Clorox® (5.25% sodium hypochlorite, The Clorox Co, US) for 60 min, once in 30% Clorox® for 30 min and then rinsed thrice by sterile distilled-water. Surface-sterilized seeds were germinated on 0.25% Phytagel®-solidified MS media (Murashige and Skoog, 1962) in a 250 mL glass jar vessel. The media were adjusted to pH 5.7 before autoclaving. Seedlings were cultured *in vitro* under condition of 25 ± 2°C ambient temperature, 60 ± 5% Relative Humidity (RH) and 60 ± 5 µmol m^{-2} s^{-1} Photosynthetic Proton Flux (PPF) provided by fluorescence lamps (TDL 36 W/84 Cool White 3350 lm, Philips, Thailand) with 16 h d^{-1} photoperiod. Fourteen-day-old rice seedlings were aseptically transferred to MS-liquid media, containing 0, 111, 222, 333 or 444 mM glucose combined with 0, 20, 40, 60 or 80 µM abscisic acid (ABA) using vermiculite as supporting material. The number of air-exchanges in the glass vessels was adjusted to 2.32 h^{-1} by punching a hole on plastic cap (⌀1 cm) and covering the hole with a microporous filter (0.20 µm of pore size). All seedlings were continuously cultured under the same conditions as during the seed germination and subsequently exposed to 342 mM NaCl for a week (Fig. 1). Physiological characteristics, soluble sugar content, root osmolarity, pigment concentration, morphological and anatomical characteristics, root length, root number, shoot height and cortex thickness were measured.

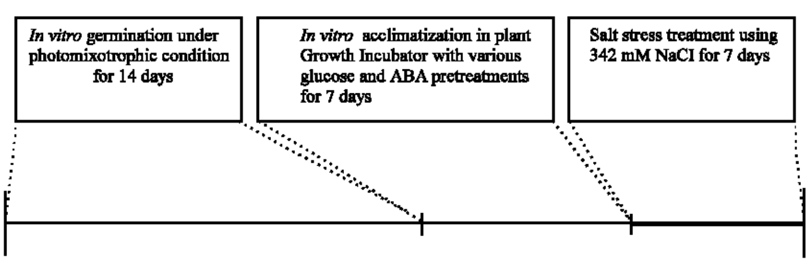

Fig. 1: Scheme of the experiment on *in vitro* photomixotrophic germination for 14 days, photoautotrophic acclimatization with various glucose and abscisic acid (ABA) pretreatments for 7 days and subsequently exposed to 342 mM NaCl for a week

Physiological Characteristics

Endogenous soluble sugars, sucrose, glucose and fructose in root organ of rice seedlings were evaluated following by Karkacier *et al*. (2003). A hundred milligram frozen material was grinded in 1.5 mL micro tube with a small pestle. One milliliter of nanopure water was added and then sonicated for 15 min. The extracted material was centrifuged at 12,000 rpm for 10 min and then the supernatant was collected. The supernatant was directly filtered through 0.45 µm pore size prior to HPLC injection. Total soluble sugar, including sucrose, glucose and fructose was analyzed using 410 differential refractometer (RI) detector and Waters 600 gradient controller pump (Waters, Milford, MA, USA) with Metacarb 87C analytical column (300×6.5 mm). Nanopure water was used as mobile phase with 0.6 mL min^{-1} flow rate. The sucrose, glucose and fructose sugar classes were used as standard.

Root osmolarity of rice seedlings was measured, according to Lanfermeijer *et al*. (1991). Fresh root tissues of rice were debris in 1.5 mL micro tube by glass rod. A twenty micro liter of extracted solution was directly dropped on a disc filter paper in osmometer chamber (Wescor, USA) and then measured.

Chlorophyll a (Chl_a), chlorophyll b (Chl_b) and carotenoid (C_{x+c}) concentrations were analyzed following the methods of Shabala *et al*. (1998) and Lichtenthaler (1987), respectively. The Chla and Chlb concentrations were measured using an UV-visible spectrophotometer (DR/4000, HACH, USA) at wavelengths 662 nm and 644 nm. The C_{x+c} concentration was measured spectrophotometrically at 470 nm. A solution of 95.5% acetone was used as a blank. The Chl_a, Chl_b and C_{x+c} (µg g^{-1} FW) concentrations in the leaf tissues were calculated according to the following equations:

$$[Chl_a] = 9.784_{662} - 0.99D^{644}$$

$$[Chl_b] = 21.42D_{644} - 4.65D_{662}$$

$$[C_{x+c}] = \frac{1000D_{470} - 1.90[Chl_a] - 63.14[Chl_b]}{214}$$

where D_i is the optical density at wavelength I.

Morphological and Anatomical Characteristics

The root length, root number and plant height of rice seedlings were measured as described by Lutts *et al*. (1996). Excised roots in size 2-3 cm from root cap were hand-sectioned and then cortex thickness was observed under light microscope (200×) (Axiostar Plus, Carl Zeiss, Germany).

Experimental Design

The experiment was designed as 5×5 factorials in Completely Randomized Design (CRD) with ten replicates and four plantlets per replication. The mean in each treatment was compared by Dancan's New Multiple Range Test (DMRT) at $p \leq 0.01$ and analyzed by SPSS software (SPSS for Windows, SPSS Inc., USA).

RESULTS AND DISCUSSION

Exogenous glucose and ABA pretreatments in the culture media of salt-stressed rice seedlings were directly enhanced on endogenous soluble sugars accumulation including sucrose, glucose and fructose. The results showed that sucrose, glucose and fructose concentration in salt-stressed root tissues were highest peak in the media supplemented with 222 mM glucose and 60 µM ABA for 86.94, 156.10 and 97.60 µg g^{-1} FW, respectively (Table 1). Sugar accumulation in salt-stressed seedlings trend to increase after exposed to low concentration of glucose (111-222 mM) and ABA

Table 1: Sucrose, glucose and fructose concentrations in the root tissues of *Indica* rice seedlings pretreated by 0. 111, 222, 333 or 444 mM glucose and 0, 20, 40, 60 or 80 µM Abscisic Acid (ABA) and subsequently exposed to 342 mM NaCl for a week

Glucose treatment (mM)	Abscisic acid (µM)	Sucrose ($\mu M\ g^{-1}$ FW)	Glucose ($\mu M\ g^{-1}$ FW)	Fructose ($\mu M\ g^{-1}$ FW)
	0	0.82h	7.71k	5.58d
	20	1.64h	20.52hij	15.55cd
0	40	3.84gh	26.71hi	23.29cd
	60	9.81efg	42.13efg	36.57bcd
	80	6.89fgh	33.85fgh	28.12cd
	0	4.12gh	15.32ij	19.05cd
	20	5.90fgh	32.46fgh	27.57cd
111	40	11.41ef	52.35de	39.72bcd
	60	21.26de	73.68c	49.30bc
	80	15.85def	61.83cd	35.77bcd
	0	6.97fgh	53.11de	29.71cd
	20	35.73c	64.86cd	38.14bcd
222	40	69.38b	100.30b	41.32bcd
	60	86.94a	156.10a	97.60a
	80	68.21b	97.43b	48.82bc
	0	4.69gh	51.26de	28.07cd
	20	6.25fgh	30.75gh	33.05cd
333	40	11.94ef	46.19ef	36.23bcd
	60	26.68cd	100.49b	72.74ab
	80	26.74cd	61.57cd	47.95bc
	0	1.97gh	10.92jk	14.00cd
	20	2.25gh	24.49hi	25.95cd
444	40	5.45fgh	33.96fgh	27.59cd
	60	18.67def	72.88c	37.55bcd
	80	8.80efg	54.91de	35.18bcd
Significant level				
Glucose		**	**	**
ABA		**	**	**
Glucose×ABA		**	**	**

Different letter(s) in each column show significant difference at $p \leq 0.01$ by Duncan's New Multiple Range Test (DMRT), Highly significant in statistics is represented by **

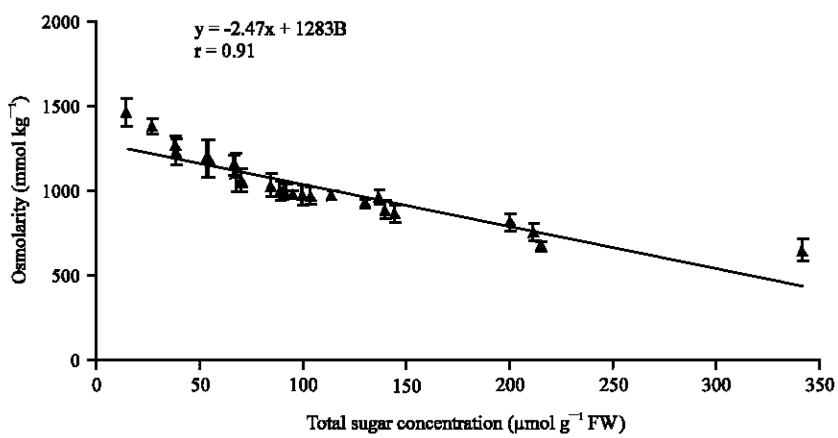

Fig. 2: Relationship between total sugar concentration and osmolarity in the root tissues of *indica* rice seedlings treated by 0. 111, 222, 333 or 444 mM glucose and 0, 20, 40, 60 or 80 µM abscisic acid (ABA) and subsequently exposed to 342 mM NaCl for a week. Error bars represent by±SE

Table 2: Chlorophyll a, chlorophyll b and total carotenoid concentrations in *Indica* rice seedlings pretreated by 0, 111, 222, 333 or 444 mM glucose and 0, 20, 40, 60 or 80 μM abscisic acid (ABA) and subsequently exposed to 342 mM NaCl for a week

Glucose treatment (mM)	Abscisic acid (μM)	Chlorophyll a ($\mu g\ g^{-1}$ FW)	Chlorophyll b ($\mu g\ g^{-1}$ FW)	Total carotenoid ($\mu g\ g^{-1}$ FW)
	0	7.54f	5.99d	9.07h
	20	14.95f	4.05d	11.34gh
0	40	21.89f	5.90d	15.18gh
	60	29.27ef	10.75cd	28.85ef
	80	9.76f	4.36d	6.51h
	0	14.52f	8.61cd	18.84gh
	20	22.19f	9.36cd	27.55ef
111	40	65.20c	17.40cd	32.91def
	60	128.05b	31.03bc	53.52bc
	80	35.51de	12.50cd	34.58de
	0	26.40ef	12.27cd	25.30ef
	20	52.64cd	14.41cd	35.54cde
222	40	141.12ab	39.58ab	57.81ab
	60	159.47a	45.00a	64.84a
	80	154.81ab	43.45a	66.46a
	0	18.22f	6.22d	15.70gh
	20	23.43ef	6.77d	23.01efg
333	40	31.13de	10.73cd	26.57ef
	60	139.78ab	45.72a	55.31ab
	80	35.59de	15.52cd	43.46bc
	0	13.12f	6.49d	15.48gh
	20	16.37f	10.43cd	18.64gh
444	40	29.29ef	12.01cd	24.56ef
	60	59.99cd	16.73cd	37.34cd
	80	34.41de	16.17cd	34.16de
Significant level				
Glucose		**	**	**
ABA		**	**	**
Glucose×ABA		**	**	**

Different letter(s) in each column show significant difference at $p \le 0.01$ by Duncan's new Multiple Range Test (DMRT), Highly significant in statistics is represented by **

(20-60 μM), while sharply drop in the high concentrations (Table 1). An endogenous sugar concentration in salt-stressed seedlings was negatively related to osmolarity in the root tissues (r = 0.91) (Fig. 2). It means that soluble sugar accumulation in salt-stressed root tissues should be played a central role as defense mechanism in osmoregulation system to be preserved the water use efficiency. A low osmolarity or water available may be directly maintained by sugar in the root organ or root zone that directly attached to salinity in the media. In aerial zone, the chlorophyll a, chlorophyll b and carotenoid concentrations in the salt-stressed leaves were maintained to the highest in 222 mM glucose and 60 μM ABA for 159.47, 45.00 and 64.84 $\mu g\ g^{-1}$ FW, respectively. The pigment concentration in salt-stressed leaves was a similar response to soluble sugar that increased in the low concentrations of glucose and ABA pretreatment and then reduced in a high dose application (Table 2). In addition to, a high osmolarity in salt-stressed roots was directly damaged on pigment concentration (r = 0.83) (Fig. 3). It should be noted that the exogenous glucose and ABA pretreatment before exposed to salt stress was an effective way to promote on soluble sugar accumulation for osmolarity control, leading to water use efficiency and pigment stabilization. The pigment stabilization in salt-stressed seedlings was positively correlated with growth performance in term of plant height (r = 0.81) (Fig. 4). The morphological and anatomical characters of salt-stressed root tissues were mentioned. Both exogenous glucose and ABA factors were directly affected on root length, root number and cortex thickness, while the glucose application does not influence on cortex thickness (Table 3). The root length, root number and cortex thickness of root organ in salt-stressed seedlings showed the highest in similar to previous pretreatment condition (222 mM glucose and 60 μM ABA) for 146.1 cm, 17.3 root and 1.3 μm, respectively (Table 3).

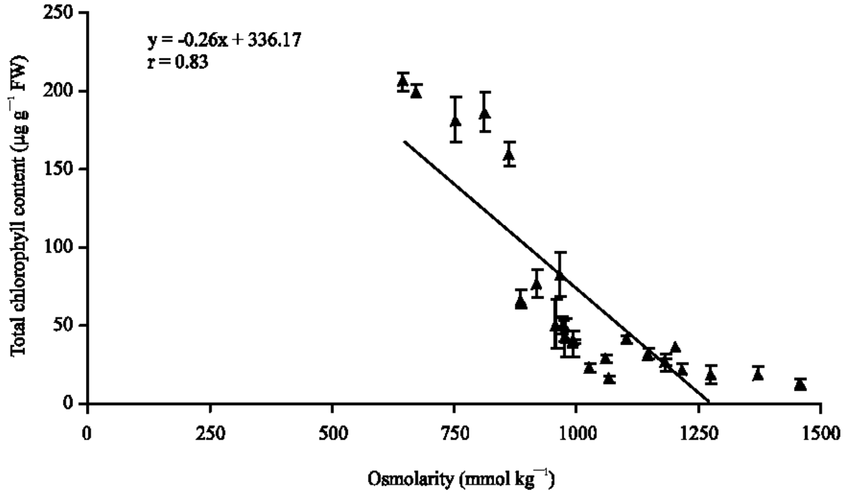

Fig. 3: Relationship between osmolarity and total chlorophyll concentration in indica rice seedlings treated by 0, 111, 222, 333 or 444 mM glucose and 0, 20, 40, 60 or 80 μM abscisic acid (ABA) and subsequently exposed to 342 mM NaCl for a week. Error bars represent by±SE

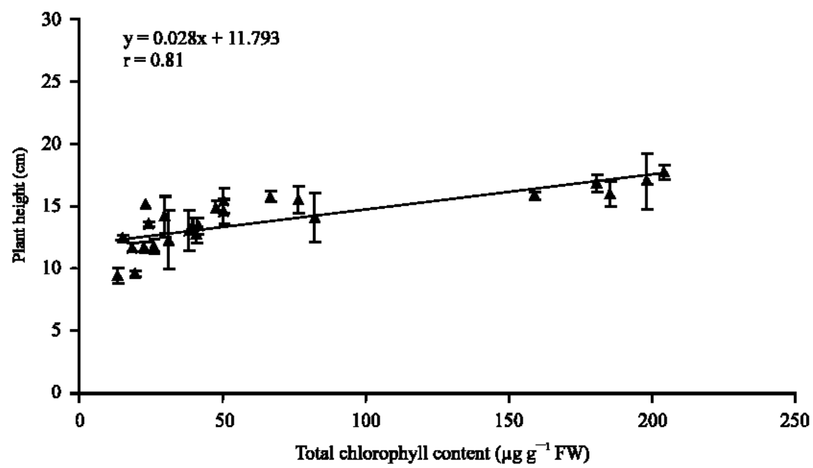

Fig. 4: Relationship between total chlorophyll concentration and plant height in indica rice seedlings pretreated by 0, 111, 222, 333 or 444 mM glucose and 0, 20, 40, 60 or 80 μM abscisic acid (ABA) and subsequently exposed to 342 mM NaCl for a week. Error bars represent by ±SE

Sugar is exogenously applied in the culture media for *in vitro* growth stimulation in higher plant species namely photomixotrophic growth (Schafer *et al*., 1992; Hdider and Desjardins, 1994; Ticha *et al*., 1998; Sima and Desjardins, 2001). Normally, the sugar concentration in the MS media is supplied on 88 mM as main carbon source for growth and development of rice crop (Cha-um *et al*., 2005) and rain tree (Mosaleeyanon *et al*., 2004). In this experiment, a high sugar concentration (111-444 mM glucose) is directly supplemented in media for osmotic regulation and energy source preservation, resulting in soluble sugar accumulation related to previous study on desert algal (Chen *et al*., 2003a) and rice crop (Cha-um *et al*., 2007). An osmolarity of the root tissues cultured on the media supplemented with a high concentration of sugar is increased, leading to plant water deficit

Table 3: Root length, root number and cortex thickness in root organ of *Indica* rice seedlings pretreated by 0, 111, 222, 333 or 444 mM glucose and 0, 20, 40, 60 or 80 μM abscisic acid (ABA) and subsequently exposed to 342 mM NaCl for a week

Glucose treatment (mM)	Abscisic acid (μM)	Root length (cm)	Root number	Cortex thickness (μm)
	0	25.3fg	9.8d	0.82cde
	20	28.4efg	11.0cd	0.88cde
0	40	29.2efg	11.8cd	0.95bc
	60	29.6efg	13.0cd	1.03abc
	80	27.5efg	11.8cd	0.85cde
	0	26.0fg	11.5cd	0.83cde
	20	36.2efg	11.8cd	1.00abc
111	40	37.1efg	12.8cd	1.08abc
	60	50.7de	14.0abc	1.13ab
	80	28.7efg	12.8cd	0.83cde
	0	46.7def	12.5cd	1.13ab
	20	82.4c	14.0abc	1.13ab
222	40	123.1b	16.8ab	1.20a
	60	146.1a	17.3a	1.30a
	80	59.7d	13.8bc	1.13ab
	0	22.3fg	10.5cd	0.75de
	20	29.5efg	12.0cd	0.88cde
333	40	30.6efg	12.0cd	0.88cde
	60	39.7def	12.8cd	1.08abc
	80	27.9efg	11.0cd	0.83cde
	0	23.0fg	10.8cd	0.63e
	20	32.8efg	10.8cd	0.75de
444	40	34.1efg	10.8cd	0.80cde
	60	34.8efg	10.8cd	0.95bc
	80	27.0efg	10.8cd	0.68de
Significant level				
Glucose		**	*	NS
ABA		**	**	**
Glucose×ABA		**	**	

**Different letters in each column show significant difference at $p \leq 0.01$ by Duncan's new Multiple Range Test (DMRT), Highly significant, significant and non-significant in statistics is represented by **, * and ^{NS}, respectively

as plant acclimatization before expose to extreme salt-stress (Chen *et al*., 2003a; Gupta and Kaur, 2005). There are many reports to mention on sugar accumulation in plant species as defensive response to salt stress in rice (Morsy *et al*., 2006), soybean (Liu and van Staden 2001), sorghum (de Lacerda *et al*., 2003; de Lacerda *et al*., 2005), kikuyu grass (Muscolo *et al*., 2003), wheat (Lutts *et al*., 2004), hexaploid triticale (Morant-Manceau *et al*., 2004), poplar (Watanabe *et al*., 2000), resurrection plant (Smith-Espinoza *et al*., 2003), salt-secretor mangrove (Parida *et al*., 2004) and citrus (Arbona *et al*., 2005). Salts, antioxidants and compatible solutes have been exogenously applied in the media to pretreated plants before exposed to salt stress (Ashraf and Foolad, 2007; Cha-um *et al*., 2006; Djanaguiraman *et al*., 2006; Cuin and Shabala, 2005; Yamane *et al*., 2004a and b). Root organ is firstly attached to salinity, which is selected and controlled the ion accumulation in plant cells using ion-pump or secrete to vacuole by ATP energy from sugar catabolism in mitochondria (Newmann *et al*., 1994; Vaughan *et al*., 2002; Bell and O'Leary, 2003; Zeng, 2005). Sugar accumulation in the plant cells is necessarily supported as energy source as well as controlled the cell osmotic pressure under soil salinity, especially root organ.

Abscisic acid or ABA is a member of endogenous hormonal regulations in higher plant, which is played an important role in signal transduction, gene(s) regulation and short-term defensive response to abiotic stresses, especially salinity (Hasegawa *et al*., 2000; Wilkinson and Davies, 2002; Seki *et al*., 2003; Sairam and Tyagi, 2004; Kaur and Gupta, 2005; Verslues and Zhu, 2005; Zhang *et al*., 2006). Generally, an ABA accumulation in plant species is progressively induced by salt stress treatment such as crop species (Degenhardt *et al*., 2000), brassica (He and Cramer, 1996), rice (Lin and Kao, 2001), maize (Jia *et al*., 2002), tomato (Chen *et al*., 2003b; Mulholland *et al*., 2003; Maggio *et al*., 2006) and barley (Fricke *et al*., 2006). Exogenous ABA treatment is an alternative way to enhance on accumulation and function as defensive response to salt stress *via* soluble sugar accumulation or osmotic adjustment in common bean (Khadri *et al*., 2006) and rice (Asch *et al*., 1995). Similarly, an ABA treatment in present study directly promotes on sucrose, glucose and fructose accumulation for osmolarity control in salt-stressed seedlings especially in the root tissues. It is similar to the previous publications that ABA applications in wheat is influence on turgor pressure and maintain on root osmolarity (Jones *et al*., 1987; Munns and Cramer, 1996), leading to root branching (Signora *et al*., 2001; de Smet *et al*., 2003; de Smet *et al*., 2006). In addition to, the endogenous sugar accumulation derived from ABA treated plants should be functioned as antioxidant (Yoshida *et al*., 2004) and reduced cell injuries (Arbona *et al*., 2006). Both sugar and ABA combinatorial functions for salinity defense mechanisms in higher plants are well established (Gazzarrini and McCourt, 2001; Knight and Knight, 2001; Ma *et al*., 2006; Rook *et al*., 2006). There is evident information on the regulation of ABA on carbohydrate metabolisms, relating to abiotic stress tolerance in term of water relation, cell/membrane stabilization, photosynthesis and overall growth performances.

In conclusion, the endogenous soluble sugar contents in the salt-stressed roots were alternatively accumulated by exogenous glucose and/or ABA application, resulting in root osmolarity control for water use efficiency in the roots as well as pigment stabilization in the leaves after exposed to salt stress. The osmolarity control and pigment stabilization in glucose and ABA pretreatment were directly stimulated on plant growth, especially the root tissues. The basic knowledge of this investigation will be further applied for rice cultivation in salinity filed trial.

ACKNOWLEDGMENTS

The authors are grateful to Dr. Teeraporn Busaya-angoon at Pathumthani Rice Research Center, for providing of Pathumthani rice seeds. This research is supported by the National Center for Genetic Engineering and Biotechnology (BIOTEC; Grant number BT-B-06-RG-14-4502).

REFERENCES

Arbona, V., A.J. Marco, Iglesias D.J. Lopez-Climent, M. Talon and A. Gomaz-Cadenas, 2005. Carbohydrate depletion in roots and leaves of salt-stressed potted *Citrus clementina* L. Plant Growth Regul., 46: 153-160.

Arbona, V., M.F. Lopez-Climent, J. Mahouachi, R.M. Perez-Clemente, S.R. Abrams and A. Gomez-Cadenas, 2006. Use of persistent analogs of abscisic acid as palliatives against salt-stress induced damage in citrus plants. J. Plant Growth Regul., 25: 1-9.

Ariyaphanphitak, W., A. Chidthaisong, E. Sarobol, V.N. Bashkin and S. Towprayoon, 2005. Effects of elevated ozone concentrations on Thai jasmine rice cultivars (*Oryza sativa* L.). Water Air Soil Poll., 167: 179-200.

Asch, F., K. Dorffling and M. Dingkuhn, 1995. Response of rice varieties to soil salinity and air humidity: A possible involvement of root-borne ABA. Plant Soil, 177: 11-19.

Ashraf, M. and M.R. Foolad, 2007. Roles of glycinebetaine and proline in improving plant abiotic stress resistance. Environ. Exp. Bot., 59: 206-216.

Bell, H.L. and J.W. O'Leary, 2003. Effects of salinity on growth and cation accumulation of *Sporobolus virginicus* (Poaceae). Am. J. Bot., 90: 1416-1424.

Cha-um, S., K. Supaibulwatana and C. Kirdmanee, 2005. Physiological responses of rice seedling (*Oryza sativa* L.) to salt -stress cultured under *in-vitro* photomixotrophic and photoautotrophic systems. Asian J. Plant Sci., 4: 136-143.

Cha-um, S., K. Supaibulwatana and C. Kirdmanee, 2006. Water relation, photosynthetic ability and growth of Thai jasmine rice (*Oryza sativa* L. spp. *indica* cv. KDML 105) to salt stress by application of exogenous glycinebetaine and choline. J. Agro. Crop Sci., 192: 25-36.

Cha-um, S., S. Roytakul, T. Sathung, A. Maijandang and C. Kirdmanee, 2007. Effect of exogenous glucose and abscisic acid on physiological and morphological performances of *in vitro* indica rice (*Oryza sativa* L. spp. *indica*). Am. J. Plant Physiol., (In Press).

Chen, L., D. Li and Y. Liu, 2003a. Salt tolerance of *Microcoleus vaginatus* Gom., a cyanobacterium isolated from desert algal crust, was enhanced by exogenous carbohydrates. J. Arid Environ., 55: 645-656.

Chen, L., X. Fu, S.H. Lips and M. Sagi, 2003b. Control of plant growth resides in the shoot and not in the root, in reciprocal grafts of *flacca* and wide-type tomato (*Lycopersicon esculentum*) in the presence and absence of salinity stress. Plant Soil, 256: 205-215.

Cuin, T.A. and S. Shabala, 2005. Exogenously supplied competible solutes rapidly ameliorate NaCl-induced potassium efflux from barley roots. Plant Cell Physiol., 46: 1924-1933.

Cushman, J.C. and H.J. Bohnert, 2000. Genomic approaches to plant stress tolerance. Curr. Opin. Plant Biol., 3: 117-124.

de Lacerda, C.F., J. Cambraia, M.A. Oliva, H.A. Ruiz and J.T. Prisco, 2003. Solute accumulation and distribution during shoot and leaf development in two sorghum genotypes under salt stress. Environ. Exp. Bot., 49: 107-120.

de Lacerda, C.F., J. Cambraia, M.A. Oliva and H.A. Ruiz, 2005. Changes in growth and in solute concentrations in sorghum leaves and roots during salt stress recovery. Environ. Exp. Bot., 54: 69-76.

de Smet, I., L. Signora, T. Beeckman, D. Inze, C.H. Foyer and H. Zhang, 2003. An abscisic acid-sensitive checkpoint in lateral root development of *Arabidopsis*. Plant J., 33: 543-555.

de Smet, I., H. Zhang, D. Inze and T. Beeckman, 2006. A novel role for abscisic acid emerges from underground. Trend Plant Sci., 11: 434-439.

Degenhardt, B., H. Gimmler, E. Hose and W. Hartung, 2000. Effect of alkaline and saline substrates on ABA contents, distribution and transport in plant roots. Plant Soil, 225: 83-94.

Demiral, T. and I. Turkan, 2006. Exogenous glycinebetaine affects growth and proline accumulation and retards senescence in two rice cultivars under NaCl stress. Environ. Exp. Bot., 56: 72-79.

Djanaguiraman, M., J.A. Sheeba, A.K. Shanker, D.D. Devi and U. Bangarusamy, 2006. Rice can acclimate to lethal level of salinity by pretreatment with sublethal level of salinity through osmotic adjustment. Plant Soil, 284: 363-373.

Flowers, T.J. and S.A. Flowers, 2005. Why dose salinity pose such a difficult problem for plant breeders? Agri. Water Manag., 78: 15-24.

Fricke, W., G. Akhiyarova, W. Wei, E. Alexandersson and A. Miller *et al*., 2006. The short-term growth response to salt of the developing barley leaf. J. Exp. Bot., 57: 1079-1095.

Gazzarrini, S. and P. McCourt, 2001. Genetic interactions between ABA, ethylene and sugar signaling pathways. Curr. Opin Plant Biol., 4: 387-391.

Ghassemi, F., A.J. Jakeman and H.A. Nix, 1995. Salinization of Land and Water Resources. University of New South Wales Press Ltd., Canberra, Australia.

Gibson, S.I., 2000. Plant sugar-response pathways. Part of a complex regulatory web. Plant Physiol., 124: 1532-1539.

Gregorio, G.B., D. Senadhira, R.D. Mendoza, N.L. Manigbas, J.P. Roxas and C.Q. Guerta, 2002. Progress in breeding for salinity tolerance and associated abiotic stresses in rice. Field Crop Res., 76: 91-101.

Gupta, A.K. and N. Kaur, 2005. Sugar signaling and gene expression in relation to carbohydrate metabolism under abiotic stresses in plants. J. Biosci., 30: 101-116.

Hasegawa, P.M., R.A. Bressan, J.K. Zhu and H.J. Bohnert, 2000. Plant cellular and molecular responses to high salinity. Ann. Rev. Plant Physiol. Plant Mol. Biol., 51: 463-499.

Hdider, C. and Y. Desjardins, 1994. Effect of sucrose on photosynthesis and phosphoenolpyruvate carboxylase activity of *in vitro* culture strawberry plantlets. Plant Cell Tiss. Org. Cult., 36: 27-33.

He, T. and G.R. Cramer, 1996. Abscisic acid concentrations are correlated with leaf area reductions in two salt-stressed rapid-cycling *Brassica* species. Plant Soil, 179: 25-33.

Jia, W., Y. Wang, S. Zhang and J. Zhang, 2002. Salt-stress-induced ABA accumulation is more sensitively triggered in roots than shoots. J. Exp. Bot., 53: 2201-2206.

Jones, H., R.A. Leigh, A.D. Tomos and R.J.W. Jones, 1987. The effect of abscisic on cell turgor pressures, solute content and growth of wheat roots. Planta, 170: 257-262.

Karkacier, M., M. Erbas, M.K. Uslu and M. Aksu, 2003. Comparison of different extraction and detection methods for sugar using amino-bonded phase HPLC. J. Chrom. Sci., 41: 331-333.

Kaur, N. and A.K. Gupta, 2005. Signal transduction pathway under abiotic stresses in plants. Curr. Sci., 88: 1771-1778.

Khadri, M., N.A. Tejera and C. Lluch, 2006. Sodium chloride-ABA interaction in two common bean (*Phaseolus vulgaris*) cultivars differing in salinity tolerance. Environ. Exp. Bot., (In Press).

Khan, M.A. and Z. Abdullah, 2003. Salinity-sodicity induced changes in reproductive physiology of rice (*Oryza sativa*) under dense soil conditions. Environ. Exp. Bot., 49: 145-157.

Khush, G.S., 2005. What it will take to feed 5.0 billion rice consumers in 2030. Plant Mol. Biol., 59: 1-6.

Knight, H. and M.R. Knight, 2001. Abiotic stress signaling pathways: Specificity and cross-talk. Trends Plant Sci., 6: 262-267.

Lafitte, H.R., A. Ismail and J. Bennett, 2004. Abiotic stress tolerance in rice for Asia: Progress and future. Proc. 4th Int. Crop Sci. Cong. 26 Sep-1 Oct 2004, Brisbane, Australia.

Lanfermeijer, F.C., J.W. Koerselman-Kooij and A.C. Borstlap, 1991. Osmosensitivity of sucrose uptake by immature pea cotyledons disappears during development. Plant Physiol., 95: 832-838.

Leohakunjit, N. and O. Kerdchoechuen, 2007. Aroma enrichment and the change during storage of non-aromatic milled rice coated with extracted natural flavor. Food Chem., 101: 339-344.

Lichtenthaler, H.K., 1987. Chlorophylls and carotenoids: Pigments of photosynthetic biomembranes. Methods Enzymol., 148: 350-80.

Lin, C.C. and C.H. Kao, 2001. Relative importance of Na^+, Cl^- and abscisic acid in NaCl induced inhibition of root growth of rice seedlings. Plant Soil, 237: 165-171.

Liu, T. and J. van Staden, 2001. Partitioning of carbohydrates in salt-sensitive and salt-tolerant soybean callus cultures under salinity stress and its subsequent relief. Plant Growth Regul., 33: 13-17.

Lutts, S., J.M. Kinet and J. Bouharmont, 1996. NaCl-induced senescence in leaves of rice (*Oryza sativa* L.) cultivars differing in salinity resistance. Ann. Bot., 78: 389-398.

Lutts, S., M. Almansouri and J.M. Kinet, 2004. Salinity and water stress have contrasting effects on the relationship between growth and cell viability during and after stress exposure in durum wheat callus. Plant Sci., 167: 9-18.

Ma, S., Q. Gong and H.J. Bohnert, 2006. Dissecting salt stress pathways. J. Exp. Bot., 57: 1097-1107.

Maggio, A., G. Raimondi, A. Martino and S. de Pascale, 2006. Salt stress response in tomato beyond the salinity tolerance threshold. Environ. Exp. Bot., (In Press).

Morant-Manceau, A., E. Pradier and G. Tremblin, 2004. Osmotic adjustment, gas exchanges and chlorophyll fluorescence of a hexaploid triticale and its parental species under salt stress. J. Plant Physiol., 161: 25-33.

Morsy, M.R., L. Jouve, J.F. Hausman, L. Hoffmann and J.M. Stewart, 2006. Alteration of oxidative and carbohydrate metabolism under abiotic stress in two rice (*Oryza sativa* L.) genotypes contrasting in chilling tolerance. J. Plant Physiol., (In Press).

Mosaleeyanon, K., S. Cha-um and C. Kirdmanee, 2004. Enhanced growth and photosynthesis of rain tree (*Samanea saman*) plantlets *in vitro* under CO_2 enrichment with decreased sucrose concentration in the medium. Sci. Hortic., 103: 51-63.

Mulholland, B.J., I.B. Taylor, A.C. Jackson and A.J. Thompson, 2003. Can ABA mediate responses of salinity stressed tomato? Environ. Exp. Bot., 50: 17-28.

Munns, R. and G.R. Cramer, 1996. Is coordination of leaf and root growth mediated by abscisic acid? Opinion. Plant Soil, 185: 33-49.

Murashige, T. and F. Skoog, 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant, 15: 473-497.

Muscolo, A., M.R. Panuccio and M. Sidari, 2003. Effects of salinity on growth, carbohydrate metabolism and nutritive properties of kikuyu grass (*Pennisetum clandestinum* Hochst). Plant Sci., 164: 1103-1110.

Ndayiragije, A. and S. Lutts, 2006. Do exogenous polyamines have an impact on the response of a salt-sensitive rice cultivar to NaCl? J. Plant Physiol., 163: 506-516.

Newmann, P.M., H. Azaizeh and D. Leon, 1994. Hardening of root cell walls: A growth inhibitory response to salinity stress. Plant Cell Environ., 16: 303-309.

Parida, A.K., A.B. Das, Y. Sanada and P. Mohanty, 2004. Effects of salinity on biochemical components of the mangrove, *Aegiceras corniculatum*. Aqua. Bot., 80: 77-87.

Rook, F., S.A. Hadingham, Y. Li and M.W. Bevan, 2006. Sugar and ABA response pathways and the control of gene expression. Plant Cell Environ., 29: 426-434.

Roy, P., K. Niyogi, D.N.S. Gupta and B. Ghosh, 2005. Spermidine treatment to rice seedlings recovers salinity stress-induced damage of plasma membrane and PM-bound H^+-ATPase in salt-tolerant and salt-sensitive rice cultivars. Plant Sci., 168: 583-591.

Sairam, R.K. and A. Tyagi, 2004. Physiology and molecular biology of salinity stress tolerance in plants. Curr. Sci., 86: 407-421.

Salekdeh, G.H., J. Siopongco, L.J. Wade, B. Ghareyazie and J. Bennett, 2002. A proteomic approach to analyzing drought-and salt-responsiveness in rice. Field Crop Res., 76: 199-219.

Schafer, C., H. Simper and B. Hofmann, 1992. Glucose feeding results in coordinated changes of chlorophyll content, ribulose-1,5-bisphosphate carboxylase-oxygenase activity and photosynthetic potential in photoautotrophic suspension cultured cells of *Chenopodium rubrum*. Plant Cell Environ., 15: 343-350.

Seki, M., Kamei A., K.Yamaguchi-Shinozaki and K. Shinozaki, 2003. Molecular responses to drought, salinity and frost: Common and different paths for plant protection. Curr. Opin. Biotech., 14: 194-199.

Senadhira, D., F.J. Zapata-Arias, G.B. Gregorio, M.S. Alejar and H.C. de la Cruz *et al*., 2002. Development of the first salt-tolerant rice cultivar through *indica*/*indica* anther culture. Field Crop Res., 76: 103-110.

Shabala, S.N., S.I. Shabala, A.I. Martynenko, O. Babourina and I.A. Newman, 1998. Salinity effect on bioelectric activity, growth, Na^+ accumulation and chlorophyll fluorescence of maize leaves: A comparative survey and prospects for screening. Aust. J. Plant Physiol., 25: 609-616.

Shannon, M.C., J.D. Rhoades, J.H. Draper, S.C. Scardaci and M.D. Spyres, 1998. Assessment of salt tolerance in rice cultivars in response to salinity problems in California. Crop Sci., 38: 394-398.

Signora, L., I. De Smet, C.H. Foyer and H. Zhang, 2001. ABA plays a central role in mediating the regulatory effects of nitrate on root branching in *Arabidopsis*. Plant J., 28: 655-662.

Sima, B.D. and Y. Desjardins, 2001. Sucrose supply enhances phosphoenolpyruvate carboxylase phosphorylation level in *in vitro Solanum tuberosum*. Plant Cell Tiss. Org. Cult., 67: 235-242.

Singh, K.N. and R. Chatrath, 2001. Salinity tolerance. In: Reynolds, M.P., J.I. Ortiz-Monasterio and A. McNab (Eds.), Application of Physiology in Wheat Breeding. Mexico, D.F.: CIMMYT.

Smith-Espinoza, C.J., A. Richter, F. Salamini and D. Bartels, 2003. Dissecting the response to dehydration and salt (NaCl) in the resurrection plant *Craterostigma plantagineum*. Plant Cell Environ., 26: 1307-1315.

Ticha, I., F. Cap, D. Pacovska, P. Hofman, D. Haisel, V. Capkova and C. Schafer, 1998. Culture on sugar medium enhances photosynthetic capacity and light resistance of plantlets grown *in vitro*. Physiol. Plant, 102: 155-62.

Vaughan, L.V., J.W. MacAdam, S.E. Smith and L. Dudley, 2002. Root growth and yield of differing alfalfa rooting populations under increasing salinity and zero leaching. Crop Sci., 42: 2064-2071.

Verslues, P.E. and J.K. Zhu, 2005. Before and beyond ABA: Upstream sensing and internal signals that determine ABA accumulation and response under abiotic stress. Biochem. Soc. Trans., 33: 375-379.

Wang, Z., B. Huang and Q. Xu, 2003. Effect of abscisic acid on drought responses of Kentucky bluegrass. J. Amer. Hortic. Sci., 128: 36-41.

Watanabe, S., K. Kojima, Y. Ide and S. Sasaki, 2000. Effects of saline and osmotic stress on proline and sugar accumulation in *Populus euphratica in vitro*. Plant Cell Tiss. Org. Cult., 63: 199-206.

Wilkinson, S. and W.J. Davies, 2002. ABA-based chemical signaling: The co-ordination of responses to stress in plants. Plant Cell Environ., 25: 195-210.

Yamane, K., M.S. Rahman, M. Kawasaki, M. Taniguchi and H. Miyake, 2004a. Pretreatment with antioxidants decreases the effects of salt stress on chloroplast ultrastructure in rice leaf segments (*Oryza sativa* L.). Plant Prod. Sci., 7: 292-300.

Yamane, K., M.S.Rahman, M. Kawasaki, M. Taniguchi and H. Miyake, 2004b. Pretreatment with a low concentration of methyl viologen decreases the effects of salt stress on chloroplast ultrastructure in rice leaf segments (*Oryza sativa* L.). Plant Prod. Sci., 7: 435-441.

Yin, C., B. Duan, X. Wang and C. Li, 2004. Morphological and physiological responses of two contrasting Poplar species to drought stress and exogenous abscisic acid application. Plant Sci., 167: 1091-1097.

Yoshida, K., E. Igarashi, E. Wakatsuki, K. Miyamoto and K. Hirata, 2004. Mitigation of osmotic and salt stresses by abscisic acid throught reduction of stress-derived oxidative damage in *Chlamydomonas reinhardtii*. Plant Sci., 167: 1335-1341.

Zeng, L. and M.C. Shannon, 2000. Salinity effects on seedling growth and yield components of rice. Crop Sci., 40: 996-1003.

Zeng, L., S.M. Lesch and C.M. Grieve, 2002. Rice growth and yield respond to changes in water depth and salinity stress. Agric. Water Manage., 59: 67-75.

Zeng, L., 2005. Exploration of relationships between physiological parameters and growth performance of rice (*Oryza sativa* L.) seedlings under salinity stress using multivariate analysis. Plant Soil, 268: 51-59.

Zhang, J., W. Jia, J. Yang and A.M. Ismail, 2006. Role of ABA in integrating plant responses to drought and salt stresses. Field Crops Res., 97: 111-119.