ACTION OF PLANT GROWTH REGULATORS IN ALLEVIATING SALINITY AND TEMPERATURE EFFECTS ON THE GERMINATION OF *PHRAGMITES KARKA*

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Abstract

High salinity, high temperature and absence of light may disturb the balance in endogenous growth regulators. This may put constraint on seed germination of halophytic grasses forcing them to adopt necessary measure like going into dormancy. Germination regulating chemicals may release dormancy imposed by such factors hence effect of Thiourea (10 mM), Nitrate (20 mM), Proline (0.1 mM), Betaine (0.1 mM), GA$_3$ (3 mM), Kinetin (0.05 mM) and Fusicoccin (5 µM) was studied in alleviating the inhibitory effect of a range of NaCl and temperature on seed germination of the halophytic grass *Phragmites karka*. Six NaCl concentrations (0, 100, 200, 300, 400 & 500 mM) were used in 12h light: 12h dark photoperiod and in complete darkness, at different temperature regimes (10/20ºC, 15/25ºC, 20/30ºC and 25/35ºC, the lower temperature corresponding to dark and the higher to light photoperiod). Highest number of seeds germinated in non-saline control and the seed germination decreased with increase in salinity at all temperature regimes. All growth regulators significantly promoted seed germination in saline and also in non-saline conditions at all thermoperiods, except at 25-35°C. Only GA$_3$ and fusicoccin successfully alleviated salinity enforced dormancy of seeds at this thermoperiod. Growth regulators promoted germination in darkness at all temperature regimes. Rate of germination was also significantly affected by the application of these chemicals. Salt induced dormancy of *P. karka* seeds was broken by the application of different growth regulators. These chemicals also alleviated the temperature (GA$_3$ and fusicoccin) and light (Nitrate) enforced dormancy from seeds of *P. karka*. It is concluded that germination regulating chemicals have differential effect on the seed germination of *P karka*. Some of them may alleviate the dormancy while others have no effect.

Introduction

Seed germination is the first and probably the most sensitive stage in the life cycle of a plant. This attains even more significance in case of halophytes which are plants of the wild occupying saline habitats. Under such conditions, increase in NaCl concentrations may inhibit the germination of seed either by creating low osmotic potential, external to the seed preventing water uptake, or through the toxic effects of Na$^+$ and Cl$^-$ ions (specific ion effect) on the metabolic processes (Khajeh-Hosseini *et al*., 2003; Kaya *et al*., 2006). In addition to salinity, germination under such conditions may face hazards like shortage (drought) or excess of water (water logging causing anaerobic condition), temperature extremes etc. (Gulzar & Khan, 2002) resulting in reduced germination. To survive and perpetuate under such harsh conditions, halophytic seeds adopt the strategy of acquiring dormancy may it be innate, enforced or induced and wait for favorable conditions. Dormancy relieving and germination regulating chemicals are useful in improving seed germination under stressful conditions and may break the dormancy as well.

The germination promoting effect of growth hormones in breaking seed dormancy has been reported in many species (Macchia *et al*., 2001; Duan *et al*., 2004; El-Keblawy *et al*., 2010; Bahrani & Pourreza, 2012). Germination regulating chemicals such as GA$_3$, kinetin, fusicoccin, proline, and betaine are reported to release dormancy (Ungar, 1977; Plyler & Proseus, 1996; Gulzar & Khan, 2001; El-Keblawy, 2012; Yarnia & Tabrizi, 2012). Compatible solutes, such as proline and betaine are known to play a role in the process of osmotic adjustment and they accumulate in plant under conditions of environmental stresses. They also act as antioxidants by scavenging OH$^-$ radical in oxidative stress (Heur, 2003; Hsu *et al*., 2003). Proline is also involved in several physiological roles, such as stabilization of proteins, preservation from heat denaturation of enzymes, while betaine stabilizes the structure of a key protein like Rubisco (Thakur & Sharma, 2005).

Phragmites karka (Retz.) Trin. ex steud., is a perennial reed distributed in Pakistan, Tropical Africa, Polynesia, northern Australia and tropical Asia. It has creeping rhizomes, tall and erect culms up to 10 m high and occurs in swamps and beside streams of both coastal and inland region. *P. karka* is used as decoration, weaving materiel, and for making musical instruments. Freshly collected seeds of *P. karka* lack innate dormancy and germinate promptly under optimum conditions (0 mM NaCl; 20/30ºC thermoperiod and 12-hour photoperiod) (Zehra & Khan, 2007). Salinity reduced germination but about 60% of the un-germinated seeds from high salt treatment (50 dS m^{-1} NaCl) could recover after salinity was removed, indicating they had salt-enforced dormancy (Zehra & Khan, 2007). Keeping in view our recent studies indicating that *P. karka* may be a potential source of biofuel (Abideen *et al*., 20011, 2012), the present study is designed to determine the effect of some dormancy relieving compounds in alleviating salinity, temperature and photoperiod effects in seeds of *P. karka*.

Materials and Methods

Inflorescences of *P. karka* were collected during autumn 2004 from Karachi university campus. Seeds were cleaned and surface sterilized with 0.85% Clorox (sodium hypochlorite) for 1 minute before germination.

Various concentrations of NaCl (0, 100, 200, 300, 400 & 500 mM) and Thiourea (10 mM), Nitrate (20 mM), Proline (0.1 mM), Betaine (0.1 mM), Gibberillin (3 mM), Kinetin (0.05 mM), Fusicoccin (5µM) were used based on optimal level promoting seed germination reported for other species (Gulzar & Khan, 2002). Germination was carried out in 50 mm diameter tight fitting plastic Petri plates with 5 ml of test solution. Four replicates of 25 seeds each were used for each treatment. Seeds were germinated in programmed incubator (Percival, Boone, USA) with a 12h light: 12h dark photoperiod (Sylvania cool white fluorescent lamps, irradiance of 25 µml m^{-3} s^{-1}, 400-700 nm) at different temperature regimes (10/20°C, 15/25°C, 20/30°C and 25/35°C). Percent germination was recorded on alternate days for 20 days. For observing response in complete darkness, Petri plates containing seeds were placed in black plastic bags and the germination was recorded at the 20th day. Rate of germination was calculated with the help of a modified Timson's germination velocity index = $\Sigma G/t$, where G is the percentage of seed germinated at 2-day intervals and t is the total germination period (Khan & Ungar, 2001a). The maximum possible values using our data were 50 (1000 seeds 20 days^{-1}). Germination data were arcsine transformed before statistical analysis using SPSS for windows release 9.0 (Anon., 1999). The differences among means were examined using three-way ANOVA and Bonferroni post-hoc test was used for significant differences between individual treatments.

Results

Four-way ANOVA of percentage germination indicated a significant (p<0.0001) effect of salinity, temperature, light, chemicals and their interactions (table
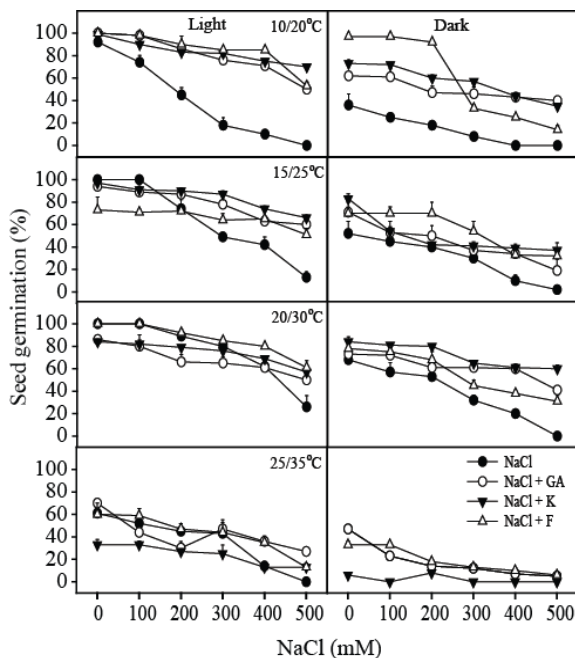
not shown). A three way ANOVA showed significant effect of salinity, temperature, chemicals and their interactions on rate of germination (table not shown). Most seeds germinated in distilled water in 12 h photoperiod, however at cooler temperature regime all seed germinated while at warmer thermoperiod only 60 % seed germinated (Fig. 1). In dark, seed germination in distilled water was also inhibited in comparison to light germinated seeds and optimal seed germination was obtained at 20/30°C (Fig. 1). Increase in salinity inhibited germination but this inhibition was greater in complete darkness. Germination was severely inhibited at 25/35°C both in light and dark while only few seeds germinated at 500 mM NaCl at moderate temperatures (20/30°C) in light (Fig. 1).

Application of GA$_3$, kinetin and fusicoccin almost completely alleviated inhibitory effect of salinity on the seeds of *P. karka* at 10/20°C (Fig. 1). Seed germination enhanced from 0 to 70% at 500 mM NaCl in kinetin and up to 60% in GA$_3$ and fusicoccin and this effect decreased with increase in temperature in both light and dark treatment (Fig. 1). Fusicoccin and GA$_3$ also alleviated temperature, light and salinity effect at 25/35°C.

Osmotica like betaine and proline almost completely alleviated salinity effect in light at 10/20°C whereas with the increase in temperature this effect of osmotica decreased and it inhibited germination at highest temperature regime used (Fig. 2). For seed germinated in dark better results were obtained at moderate temperature regimes for both osmotica and partial alleviation was noticed at coolest regime and inhibitory effect at highest temperature regime (Fig. 2).
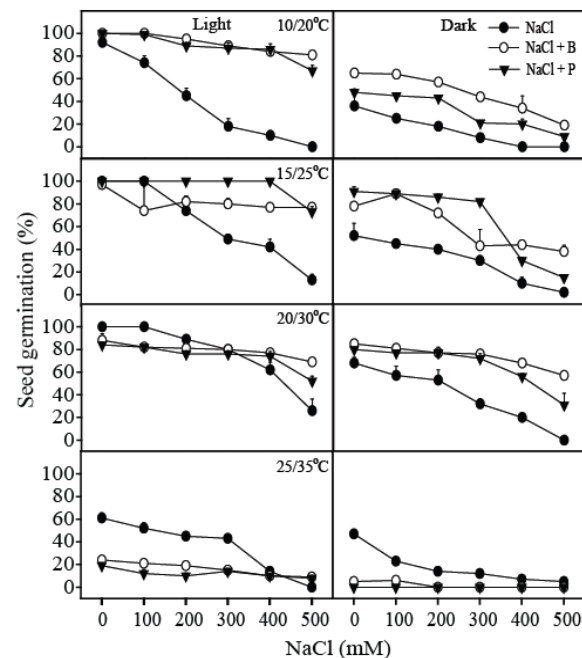

Fig. 1. Effect of plant hormones (GA$_3$, 3 mM; Kinetin, 0.05 mM and Fusicoccin, 5 µM) on final seed germination (n = 4; mean ± s.e.) of *P. karka* under various salinities (0 to 500 mM NaCl) and photoperiods (Light: 12/12 h light/dark; Dark: 24 h dark).

Fig. 2. Effect of plant osmotica (Betaine, 0.1 mM and Proline, 0.1 mM) on final seed germination (n = 4; mean ± s.e.) of *P. karka* under various salinities (0 to 500 mM NaCl) and photoperiods (Light: 12/12 h light/dark; Dark: 24 h dark).

Nitrogenous compounds like thiourea and nitrate also partially to completely alleviated the salinity effects on the seed germination of *Phragmites karka* particularly at lower temperature regimes. With the increase in temperature there was no effect at $20/30^{\circ}C$ with substantial inhibition at $25/35^{\circ}C$ (Fig. 3). Similar trends were noted in dark germinated seeds however nitrate partially alleviated salinity effect at $20/30^{\circ}C$.
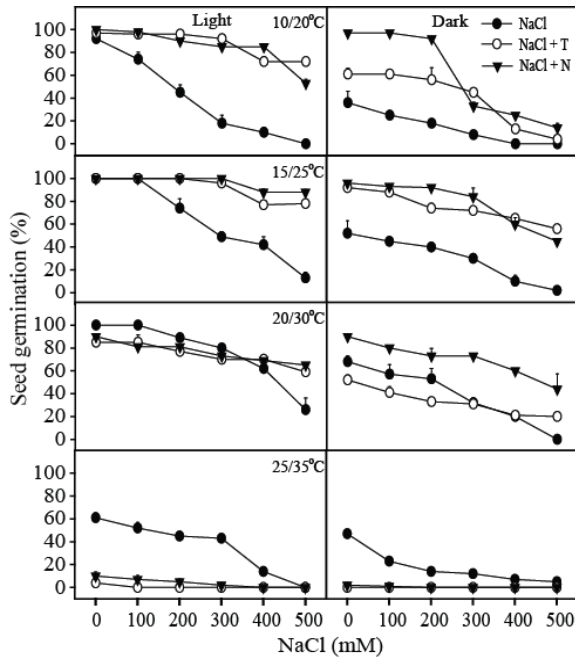

Fig. 3. Effect of nitrogenous compounds (Thiourea, 10 mM and Nitrate, 20 mM) on final seed germination (n = 4; mean ± s.e.) of *P. karka* under various salinities (0 to 500 mM NaCl) and photoperiods (Light: 12/12 h light/dark; Dark: 24 h dark).

The effect of dormancy-regulating chemicals mentioned above on the rate of seed germination more or less followed the similar pattern as that of percent seed germination (Tables 1-3). Germination regulating chemicals did not improve the rate of germination under non-saline conditions except for GA_3, kinetin, betaine and proline at $10/20^{\circ}C$. However, rate of germination under saline conditions was improved in all treatments at cooler temperature. At the higher temperature regimes of $15/25^{\circ}C$ and $20/30^{\circ}C$ the rate of germination was substantially improved at higher salinity concentrations except for fusicoccin (Tables 1-3). At warmer thermoperiod, magnitude of the improvement was much lower as compared to other temperature regimes.

Discussion

Seeds of *Phragmites karka* showed above 90% germination in non-saline control and were inhibited with an increase in salinity and no seed germinated at 500mM NaCl. Fusicoccin, GA_3, kinetin, betaine, nitrate and thiourea partially to completely alleviated the inhibitory effects of salinity on the germination. These chemicals also compensated for the light need during seed germination. These effects of dormancy-regulating chemicals are greater at cooler thermoperiod while at $25/35^{\circ}C$ seed germination was inhibited by dormancy-regulating chemicals treatment.

These chemical also improved seed germination prevented by cooler temperature.

Fusicoccin alleviated salinity induced dormancy in *P. karka*. Similar effect was also reported for the seeds of other halophytes (Gul & Khan, 2003; Khan *et al*., 2004; El-Keblawy *et al*., 2010; El-Keblawy, 2012). Fusicoccin may act as gibberellic acid to counter the effects of abscisic acid produced due to salinity effects. This alleviation may be due to the stimulation of ATPase production to facilitate proton extrusion and K^+ uptake (Antipova *et al*., 2003). Fusicoccin is known to promote seed germination by the stimulation of cell elongation and cell division in germinating seeds (Obroucheva & Antipova, 1992). Exogenous fusicoccin promoted radical emergence by inducing initiation of cell elongation in hypocotyls and in radical of *Vicia faba* seeds (Antipova *et al*., 2003).

Proline and betaine are reported to partially alleviate innate dormancy in *Zygophyllum simplex*, *Atriplex stocksii*, *A. prostrata*, *Halogeton glomeratus,* and *Halopyrum mucronatum* (Khan & Ungar, 1997; Khan *et al*., 1998; Khan & Ungar, 2000, 2001c; Khan *et al*., 2003). However, proline and betaine did not relieve salinity-induced dormancy in *Arthrocnemum indicum* (Khan *et al*., 1998) *Kosteletzkya virginica* (Poljakoff-Mayber *et al*., 1994), *Halopyrum mucronatum* (Khan & Ungar, 2001c), *Salicornia rubra* (Khan *et al*., 2002) and *Salicornia utahensis* (Gul & Khan, 2003). Both proline and betaine alleviated the innate dormancy of *Zygophyllum simplex* seeds but neither was effective at high salinities (Khan & Ungar, 1997) while Gul & Weber (1998) reported that proline and betaine both alleviated high salinity effects in *Allenrolfea occidentalis*. Our results with *P. karka* showed that both betaine and proline were very effective in alleviating salinity induced dormancy.

Thiourea and nitrate both stimulated the germination of *P. karka* seeds under saline conditions. Gul & Weber (1998) reported that nitrate and thiourea almost completely alleviated the seed germination of *Allenrolfea occidentalis* at all salinities used. A substantial promotion was also reported in *Triglochin maritima, Salicornia utahensis, Halogeton glomeratus, Sporobolus arabicus* and *Aeluropus lagopoides* (Khan & Ungar, 1999, 2001a,b; Gul & Khan, 2003; Gulzar & Khan, 2002). Some nitrogenous compounds such as nitric oxide, nitrate, nitrite and thiourea are known to stimulate the germination of seeds (Bethke *et al*., 2004; Li *et al*., 2005). Thiourea is also known to break dormancy and overcome the negative effect of temperature on seed germination (Esashi *et al*., 1979; Aldosaro *et al*., 1981).

Seed germination of *P. karka* was partially alleviated by the application of GA_3 and kinetin. The GA_3 is known to alleviate salinity effect in some halophytic seeds (Khan & Ungar, 1998; Khan *et al*., 1998; Li *et al*., 2005) while it was ineffective in other halophytes like *Suaeda fruticosa* and *Haloxylon recurvum* (Khan & Gul, 2006), *Sarcobatus vermiculatus* (Gul *et al*., 2001), *Ceratoides lanata* (Khan *et al*., 2004). Gibberellic acid was also found to be ineffective in alleviating salinity effects on the germination of number sub-tropical halophytes (Khan & Gul, 2006) however, it alleviated salinity effects in case of *Zygophyllum simplex* (Khan & Ungar, 1999) and *Halopyrum mucorantum* (Khan & Ungar, 2001c). Kinetin is also a more potent growth regulator known to alleviate salinity effects in a number of halophytes (Gul & Khan, 2003; Khan *et al*., 2002, 2004; Li *et al*., 2005; Khan & Gul, 2006).

Table 1. Effect of plant hormones (GA₃, 3 mM; Kinetin, 0.05 mM and Fusicoccin, 5 µM) on rate of seed germination (n = 4; mean ± s.e.) of *P. karka* under various salinities (0 to 500 mM NaCl) and 12/12 h light/dark photoperiod (Similar letters within each NaCl concentration indicate non-significant differences between treatments by Bonferroni test at *p*<0.05.

	Salinity (mM)					
	0	**100**	**200**	**300**	**400**	**500**
10/20 °C						
NaCl	32 ± 2.69ᵃ	23 ± 2.23ᵇ	11 ± 1.25ᶜ	5 ± 1.73ᶜ	2 ± 0.80ᵇ	0 ± 0.00ᶜ
GA₃	38 ± 0.60ᵃ	37 ± 0.26ᵃ	34 ± 3.00ᵃ	30 ± 0.64ᵃ	27 ± 0.95ᵃ	20 ± 1.38ᵃ
Kinetin	40 ± 0.64ᵃ	36 ± 0.76ᵃ	34 ± 0.41ᵃ	33 ± 0.42ᵃ	28 ± 0.92ᵃ	27 ± 1.52ᵃ
Fusicoccin	30 ± 4.84ᵃ	29 ± 4.53ᵇ	27 ± 2.97ᵇ	23 ± 1.95ᵇ	20 ± 1.15ᵃ	13 ± 2.26ᵇ
15/25 °C						
NaCl	41 ± 0.08ᵃ	40 ± 0.51ᵃ	27 ± 2.30ᵇ	16 ± 1.15ᶜ	13 ± 1.87ᵇ	3 ± 1.18ᶜ
GA₃	36 ± 2.49ᵃᵇ	35 ± 3.30ᵃᵇ	34 ± 0.86ᵃ	30 ± 0.54ᵃ	23 ± 1.72ᵃ	23 ± 1.45ᵃ
Kinetin	35 ± 1.49ᵇ	32 ± 1.64ᵇ	32 ± 0.94ᵃ	31 ± 1.98ᵃ	27 ± 1.46ᵃ	23 ± 0.76ᵃ
Fusicoccin	29 ± 4.39ᶜ	28 ± 0.78ᶜ	27 ± 1.56ᵇ	26 ± 3.17ᵇ	25 ± 0.70ᵃ	19 ± 2.30ᵇ
20/30 °C						
NaCl	46 ± 1.41ᵃ	43 ± 0.83ᵃ	35 ± 1.05ᵃᵇ	26 ± 0.51ᵇ	19 ± 1.33ᶜ	7 ± 1.90ᵇ
GA₃	34 ± 4.22ᵇ	34 ± 2.14ᵇ	28 ± 2.63ᶜ	27 ± 0.75ᵇ	25 ± 1.02ᵇ	20 ± 2.14ᵃ
Kinetin	35 ± 0.65ᵇ	33 ± 2.47ᵇ	32 ± 0.94ᵇᶜ	30 ± 2.71ᵃ	26 ± 0.51ᵇ	22 ± 0.71ᵃ
Fusicoccin	42 ± 0.70ᵃ	41 ± 0.42ᵃ	38 ± 0.57ᵃ	35 ± 1.70ᵃ	33 ± 0.17ᵃ	24 ± 3.56ᵃ
25/35 °C						
NaCl	46 ± 1.41ᵃ	43 ± 0.83ᵃ	35 ± 1.05ᵃ	26 ± 0.51ᵃ	19 ± 1.33ᵇ	7 ± 1.90ᵇ
GA₃	34 ± 4.22ᵇ	34 ± 2.14ᵇ	28 ± 2.63ᵇ	27 ± 0.75ᵃ	25 ± 1.02ᵃ	20 ± 2.14ᵃ
Kinetin	35 ± 0.65ᵇ	33 ± 2.47ᵇ	32 ± 0.94ᵃ	30 ± 2.71ᵃ	26 ± 0.51ᵃ	22 ± 0.71ᵃ
Fusicoccin	22 ± 2.87ᶜ	21 ± 2.48ᶜ	18 ± 1.09ᶜ	16 ± 4.53ᵇ	13 ± 2.00ᶜ	4 ± 1.12ᵇ

Table 2. Effect of plant osmotica (Betaine, 0.1 mM and Proline, 0.1 mM) on rate of seed germination (n = 4; mean ± s.e.) of *P. karka* under various salinities (0 to 500 mM NaCl) and 12/12 h light/dark photoperiod (Similar letters within each NaCl concentration indicate non-significant differences between treatments by Bonferroni test at *p*<0.05.

	Salinity (mM)					
	0	**100**	**200**	**300**	**400**	**500**
10/20 °C						
NaCl	32 ± 2.69ᵇ	23 ± 2.23ᵇ	11 ± 1.25ᵇ	5 ± 1.73ᵇ	2 ± 0.80ᵇ	0 ± 0.00ᶜ
Betaine	40 ± 0.83ᵃ	38 ± 1.14ᵃ	36 ± 1.28ᵃ	33 ± 1.44ᵃ	30 ± 1.42ᵃ	29 ± 1.19ᵃ
Proline	41 ± 0.22ᵃ	40 ± 0.57ᵃ	38 ± 0.33ᵃ	34 ± 0.89ᵃ	31 ± 1.28ᵃ	23 ± 0.97ᵇ
15/25 °C						
NaCl	41 ± 0.08ᵃ	40 ± 0.51ᵃ	27 ± 2.30ᵇ	16 ± 1.15ᵇ	13 ± 1.87ᶜ	3 ± 1.18ᵇ
Betaine	39 ± 0.33ᵃ	34 ± 1.70ᵇ	32 ± 1.93ᵃᵇ	29 ± 3.38ᵃ	26 ± 0.87ᵇ	27 ± 0.81ᵃ
Proline	42 ± 1.14ᵃ	41 ± 1.24ᵃ	38 ± 1.21ᵃ	35 ± 0.41ᵃ	32 ± 0.81ᵃ	24 ± 2.50ᵃ
20/30 °C						
NaCl	46 ± 1.41ᵃ	43 ± 0.83ᵃ	35 ± 1.05ᵃ	26 ± 0.51ᵇ	19 ± 1.33ᵇ	7 ± 1.90ᶜ
Betaine	38 ± 2.64ᵇ	34 ± 0.49ᵇ	33 ± 0.46ᵃ	33 ± 0.44ᵃ	31 ± 1.26ᵃ	26 ± 1.07ᵃ
Proline	33 ± 0.61ᵇ	31 ± 0.73ᵇ	29 ± 0.63ᵇ	27 ± 1.02ᵇ	27 ± 1.19ᵃ	18 ± 1.64ᵇ
25/35 °C						
NaCl	46 ± 1.41ᵃ	43 ± 0.83ᵃ	35 ± 1.05ᵃ	26 ± 0.51ᵇ	19 ± 1.33ᶜ	7 ± 1.90ᶜ
Betaine	38 ± 2.64ᵇ	34 ± 0.49ᵇ	33 ± 0.46ᵃ	33 ± 0.44ᵃ	31 ± 1.26ᵃ	26 ± 1.07ᵃ
Proline	33 ± 0.61ᵇ	31 ± 0.73ᵇ	29 ± 0.63ᵇ	27 ± 1.02ᵇ	27 ± 1.19ᵇ	18 ± 1.64ᵇ

Table 3. Effect of nitrogenous compounds (Thiourea, 10 mM and Nitrate, 20 mM) on rate of seed germination (n = 4; mean ± s.e.) of *P. karka* under various salinities (0 to 500 mM NaCl) and 12/12 h light/dark photoperiod (Similar letters within each NaCl concentration indicate non-significant differences between treatments by Bonferroni test at *p*<0.05.

	Salinity (mM)					
	0	**100**	**200**	**300**	**400**	**500**
10/20 °C						
NaCl	32 ± 2.69[a]	23 ± 2.23[a]	11 ± 1.25[b]	5 ± 1.73[c]	2 ± 0.80[b]	0 ± 0.00[b]
Thiourea	25 ± 0.06[b]	24 ± 0.81[a]	23 ± 0.49[a]	19 ± 0.47[b]	18 ± 1.36[a]	17 ± 0.61[a]
Nitrate	32 ± 1.52[a]	28 ± 1.02[a]	25 ± 0.39[a]	23 ± 0.45[a]	21 ± 0.41[a]	15 ± 0.36[a]
15/25 °C						
NaCl	41 ± 0.08[a]	40 ± 0.51[a]	27 ± 2.30[b]	16 ± 1.15[c]	13 ± 1.87[b]	3 ± 1.18[c]
Thiourea	41 ± 0.59[a]	40 ± 0.19[a]	40 ± 0.10[a]	38 ± 0.54[b]	30 ± 1.30[a]	29 ± 0.46[b]
Nitrate	44 ± 0.48[a]	44 ± 0.34[a]	44 ± 0.43[a]	42 ± 0.43[a]	34 ± 0.74[a]	34 ± 0.62[a]
20/30 °C						
NaCl	46 ± 1.41[a]	43 ± 0.83[a]	35 ± 1.05[a]	26 ± 0.51[b]	19 ± 1.33[b]	7 ± 1.90[b]
Thiourea	34 ± 2.18[b]	34 ± 1.39[b]	30 ± 1.14[a]	28 ± 1.37[ab]	28 ± 0.53[a]	22 ± 0.44[a]
Nitrate	37 ± 2.55[b]	34 ± 0.54[b]	34 ± 0.59[a]	31 ± 1.37[a]	30 ± 0.97[a]	27 ± 0.31[a]
25/35 °C						
NaCl	46 ± 1.41[a]	43 ± 0.83[a]	35 ± 1.05[a]	26 ± 0.51[b]	19 ± 1.33[b]	7 ± 1.90[b]
Thiourea	34 ± 2.18[b]	34 ± 1.39[b]	30 ± 1.14[a]	28 ± 1.37[ab]	28 ± 0.53[a]	22 ± 0.44[a]
Nitrate	37 ± 2.55[b]	34 ± 0.54[b]	34 ± 0.59[a]	31 ± 1.37[a]	30 ± 0.97[a]	27 ± 0.31[a]

Change in temperature also influenced the germination of *P. karka* showing inhibition at low and high temperature regimes. Temperature stress may inhibit seed germination through ABA biosynthesis and catabolism (Goani *et al*., 2004). Low temperature (10/20°C) induced dormancy was broken by applying different growth regulators, but at high temperature regime (25/35°C) only GA_3 and fusicoccin alleviated the dormancy of seeds. There is no reported information about the effect of growth regulators in breaking temperature stress induced dormancy from seeds of halophytes.

Germination of many halophytes occurs when there is an optimal combination of day length, thermoperiod and salinity (Young *et al*., 1980). Seeds of *P. karka* showed a light requirement for germination, they were able to germinate under light at high percentages even in salinity, while its germination was inhibited in complete darkness both in saline and non-saline conditions. Nitrate promoted germination or broke dark enforced dormancy of *P. karka* seeds from lower to moderate thermopreiods (10/30°C). Nitrate has been commonly used to break dormancy in seeds requiring light to germinate (Baskin & Baskin, 1998). For instance, seed germination of *Hypericum*. Spp that is affected by the absence of light was enhanced by KNO_3 application (Cirak *et al*., 2007).

Phragmites karka which occupies a relatively wetter part of the coastal and inland areas around Karachi is highly tolerant to salinity at germination stage. In the present study its seeds exhibited dormancy in stress conditions and had a requirement of growth regulators to break this dormancy to allow seeds to germinate. Application of proline, betaine, thiourea, nitrate, fusicoccin, GA_3, and kinetin substantially alleviated the salinity effects on germination. The germination regulating chemicals were most effective at cooler temperature regime. Dark induced germination inhibition was also partially alleviated by the application of these chemicals. Higher temperature regimes decreased germination substantially where only GA_3 and fusicoccin had some alleviating effects. *Pharagmites karka* could produce plenty of biomass in a waterlogged saline soil which may be used to make bio-fuel, fodder, thatching material, baskets etc. This species could successfully be planted after monsoon rains and seedling emergence could be enhanced by priming the seed with various germination regulating chemicals.

Acknowledgement

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