Potasssium chloride as a nutrient seed primer to enhance salt-tolerance in maize

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Abstract – The objective of this work was to determine if KCl could be a useful nutrient primer for safe seed germination in maize crop under salt stress conditions. Seed priming was done using 50 mmol L^{-1} of muriate of potash, and germination and seedling growth were evaluated after salt stress with NaCl up to 50 mmol L^{-1}. Another set of seeds was tested under the same salt stress conditions without priming. Under salinity stress, germination percentage, germination rate index, germination coefficient, and seedling vigor indexes were higher in primed seeds. In unprimed seeds, mean germination time increased, while the germination rate index and the fresh and dry matter mass decreased more sharply with salinity stress. The Na/K ratio was higher in unprimed seeds.

Index terms: *Zea mays*, muriate of potash, osmopriming, saline stress, sodium chloride.

Cloreto de potássio como condicionador osmótico nutritivo de sementes para aumentar a tolerância ao sal em milho

Resumo – O objetivo deste trabalho foi determinar se o KCl pode constituir um condicionador osmótico nutritivo, para a germinação segura da cultura do milho em condições de estresse salino. O osmocondicionamento das sementes foi feito com 50 mmol L^{-1} de muriato de potássio, e a germinação e o desenvolvimento de plântulas foram avaliados após estresse salino por NaCl, até 50 mmol L^{-1}. Outro lote de sementes foi testado nas mesmas condições de estresse salino, sem condicionamento. Em condições de estresse salino, a percentagem de germinação, o índice da taxa de germinação, o coeficiente de germinação e os índices de vigor das plântulas foram maiores nas sementes condicionadas. Em sementes não condicionadas, o tempo médio de germinação aumentou, e o índice da taxa de germinação e as massas de matéria fresca e seca diminuíram mais acentuadamente com o estresse salino. A relação Na/K foi maior nas sementes não condicionadas.

Termos para indexação: *Zea mays*, muriato de potássio, osmocondicionamento, estresse salino, cloreto de sódio.

The osmotic effect of salts in the soil may restrict seed germination processes. The ionic status of a salt is relative, depending on the glycophytic or halophytic nature of the plant species. A wide range of soluble ions in the soil solution is permissible for glycophytes. The presence of sodium ions in the soil solution is acceptable by such plants, but only in a limited range, being harmful above a threshold limit.

The salinity-tolerance of maize is very limited, and high-salt stress alters its growth responses (Chartzoulakis & Klapaki, 2000), especially in seedlings, which may be less tolerant to salt-stress than adults (Geraldine & Donovan, 1999). However, the abnormal effects of salinity stress on germination can be diminished by various seed priming agents (Roy & Srivastava, 2000). Halo-priming of seeds in pre-sowing treatments in an osmotic solution allows seeds to absorb water, but restricts radicle occurrence through testa until the primed seeds are sown for germination under salt stress conditions. Primed seeds usually show improved germination parameters (Hardegree & Van Vactor, 2000). Nutrient priming has the additive advantage of improving K supply to plants (Al-Mudaris & Jutzi, 1999).

Potassium chloride is the most widely used source of potassium for agricultural crops, and Cl is considered an essential micronutrient for optimal growth (Fixen, 1993). Potassium chloride has been introduced as the osmoticum to enhance germination, emergence and growth of Poaceae plants (Misra & Dwivedi, 1980).

The objective of this work was to determine if KCl could be a useful nutrient primer for safe seed germination in maize crop under salt stress conditions.

The experiment was carried out at the Salinity Laboratory of the Land Resources Research Institute, National Agriculture Research Center, at 33°42' N, 73°08' E, at 518 m altitude in Islamabad, Pakistan.

Two hundred and fifty seeds of *Zea mays* L., cultivar Gold, were surface-sterilized with 1% sodium hypochlorite for 15 min (Britto & Kronzucker, 2002). Seeds were then washed three times with distilled water. For halopriming, seeds were placed in a 250 mL beaker and a solution with 50 mmol L^{-1} potassium chloride was applied. Seeds were kept in the solution for five hours and were then surface-dried with tissue paper, and air-dried for six hours. Ten seeds were placed on filter paper in Petri dishes (11 cm diameter), in quadruplicate, for germination under salinity stress.

To impose salt stress, sodium chloride was used at 0, 20, 30, 40 and 50 mmol L^{-1}, dissolved in the nutrient solution composition (Hoagland & Arnon, 1950). Petri dishes were covered and placed in the dark at 25°C, for germination. Germinated seeds were counted from the 3rd to 7th day.

Mean germination time (MGT) was calculated according to the equation of Ellis & Roberts (1980). Observed germination percentage was arcsin transformed for statistical analysis, as proposed by Dezfuli et al. (2008). The germination rate index (GRI) was calculated as given by Islam et al. (2000). Seedling vigor index (VIG) was calculated as given by Karaguzel et al. (2004). The germination coefficient (GC) was determined according to Copeland (1976). After recording fresh matter mass (FM), the seedlings were rinsed with deionized water, and plant samples were dried at 65°C. After recording dry matter mass (DM), each sample was cut into small pieces, mixed, and separately digested in a perchloric-nitric (1:2) di-acid mixture (Chapman & Pratt 1961). Sodium and potassium ions in the digested material were determined by atomic absorption spectroscopy using Perkin Elmer (Waltham, MA, USA) model 4000.

Data were statistically analyzed according to two variation factors (priming treatments and NaCl doses), and means were compared using the LSD test (Gomez & Gomez, 1984).

Pre-germination of seeds with MOP and the imposed salt stress significantly affected ($p<0.01$) seed germination, seedling growth and ionic ratios.

Germination percentage (GP) was affected by salt stress. In general, GP for primed and unprimed seeds decreased with salinity stress. For primed seeds, GP was 9, 15, 19 and 25% higher than for unprimed seeds at 20, 30, 40 and 50 mmol L^{-1} NaCl stress, respectively (Table 1). Moreover, for primed and unprimed seeds, MGT increased with salinity stress. From control to 40 mmol L^{-1} NaCl application, MGT remained unaffected. However, at 50 mmol L^{-1} NaCl stress, MGT for primed seeds was lower than that of unprimed ones. Germination increases as MGT decreases (Sedghi et al., 2010). Priming also reduced the adherence of the seed coat due to imbibition, which may permit the emergence of the radicle without resistance (Nascimento & West, 1998). Liu et al. (2002) found that halopriming, especially with NaCl, increased the activity of superoxide dismutase (SOD) and peroxidase; and by increasing the respiration rate, the germination percentage was improved. With osmopriming using KCl, GP was enhanced due to physicochemical activities in maize seeds resulting in MGT reduction.

With increasing salt stress, the GRI decreased both in unprimed and primed seeds. Moreover, GRI was higher in primed seeds than in unprimed ones. This index expresses the speed of germination; it is positively correlated to higher and faster germination, and it can be used to differentiate treatments. With increasing salinity, GRI decreased, which is similar to the results of Omar et al. (2008).

Souhail & Chaâbane (2009) reported that the germination coefficient is an indicator of the level of the germination variability. In the present study, GC decreased with increasing salinity both in unprimed and primed seeds (Table 1). Up to 40 mmol L^{-1} NaCl, the GC values were similar, but at 50 mmol L^{-1}, the GC decreased in both primed and unprimed treatments. However, at 50 mmol L^{-1}, GC was 17% higher in the primed seeds. Therefore, at 50 mmol L^{-1}, GC proved to be a good treatment discriminator.

With increasing salt stress, VIG decreased both in unprimed and primed seeds, but more acutely in unprimed ones. At 20, 30, 40 and 50 mmol L^{-1} NaCl, VIG was 13, 35, 38 and 54% lower in the unprimed seeds, respectively (Figure 1). Seedling vigor expresses seed quality, besides genetic aspects.

For primed and unprimed seedlings, FM also decreased with salinity stress. For primed seeds, however, FM was 13, 10, 6, and 6% higher than for unprimed ones at 20, 30, 40 and 50 mmol L^{-1} NaCl, respectively. Similarly, DM of both primed or unprimed seedlings decreased with salinity stress, and once more, for primed seeds, DM was 4, 5, 11, and 18% higher than for unprimed ones. Tissue development is dependent on water retention, and this phenomenon is very important under salt stress (Badr-uz-Zaman, 2010). The declining trend observed in FM and DM with increasing salt stress, which was also observed by Badr-uz-Zaman et al. (2006, 2010), is possibly due to salt effects on metabolic processes, by reducing water potential. Okçu et al. (2005) reported that reduction in seedling growth under saline conditions is due to increased sodium chloride toxicity, and to accumulation of sodium ions in the photosynthetic tissues (Davenport et al., 2005).

In primed and unprimed seedlings, the Na/K ratio increased with salinity stress. In seedlings of primed seeds, the Na/K ratio was 20 and 15% lower than in unprimed ones at 20–30 mmol L^{-1}, and 40–50 mmol L^{-1}, respectively. The increased Na/K

Table 1. Effect of NaCl application on the germination percentage, mean germination time, germination rate index and germination coefficient of maize seeds primed with KCl[(1)].

NaCl	Germination percentage			Mean germination time (Days)			Germination rate index			Germination coefficient (%)		
(mmol L^{-1})	Unprimed	Primed	Mean	Unprimed	Primed	Mean	Unprimed	Primed	Mean	Unprimed	Primed	Mean
0	56.8	56.8	56.8A	4.9	4.9	4.9D	71.4	71.4	71.4A	20.3	20.3	20.3A
20	50.8	55.2	53.0B	5.0	4.9	5.0C	64.7	68.0	66.4B	20.0	20.2	20.1B
30	45.0	51.8	48.4C	5.0	5.0	5.0C	57.1	60.0	58.6C	19.8	20.0	19.9C
40	41.2	49.2	45.2D	5.1	5.1	5.1B	50.0	53.3	51.7D	19.6	19.7	19.7D
50	36.4	45.4	40.9E	6.0	5.3	5.7A	38.3	42.0	40.2E	14.3	16.7	15.5E
Mean	24.5b	51.7a		56.3b	58.9a		56.3b	58.9a		18.8b	19.4a	

[(1)]Means followed by equal letters do not differ significantly at 1% probability.

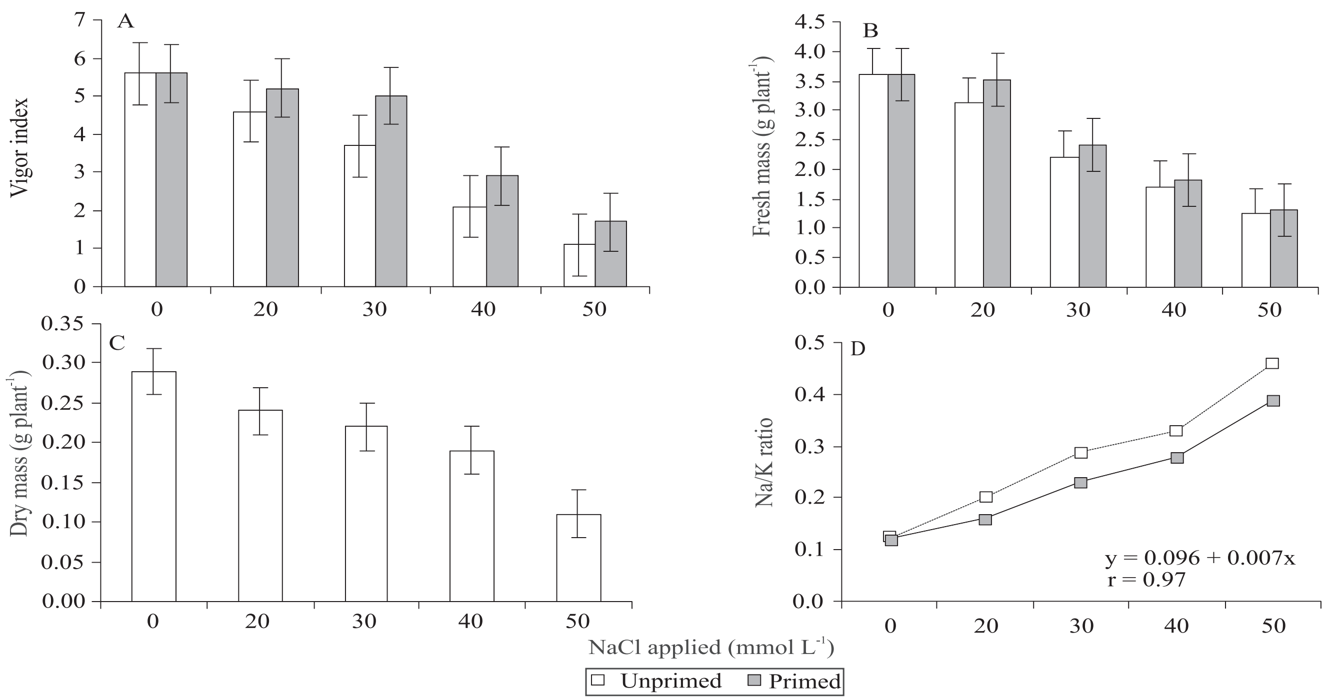

Figure 1. Effect of KCl priming of maize seeds on A, vigor index, B, fresh and C, dry matter mass, and on D, Na/K ratio of maize seedlings under NaCl stress.

ratio had a pronounced effect on plant DM, decreasing it. The increased level of Na^+ in the tissue disturbs the normal cellular function of plant; Chinnusamy et al. (2005) reported that a low Na/K ratio in the cytosol is essential for normal cell functions.

Priming with KCl can be a useful strategy to activate germination of maize seeds, leading to safe seedling stands under salt stress, since germination percentage, germination rate index, germination coefficient and seedling vigor indexes were improved through the this priming process.

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Received on February 22, 2012 and accepted on July 23, 2012