

Different amendments application in saline-sodic soils showed long-term effects on improving growth and yield of rice (*Oryza sativa* L.)

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Background. Saline-sodic soils is widely distributed in arid and semi-arid regions around the world. High level of salt and sodium concentrations inhibited the growth and development of crop. However, there has been limited report on both osmotic potential in soil solution (OP_{ss}) and characteristics of Na^+ and K^+ absorption in rice under amendments application.

Methods. A field experiment was conducted between 2009 and 2017 to analyze the influence of amendments addition in saline-sodic soils on rice growth and yield. Rice was grown in the soil with no amendment (CK), with desulfurization gypsum (DG), with sandy soil (SS), with farmyard manure (FM) and with the mixture of above amendments (M), respectively. The osmotic potential in soil solution, the selective absorption (SA), selective transport (ST), the distribution of K^+ and Na^+ and yield components in rice plants were investigated. **Results.** The results indicated that amendments application have positive effects on rice yield, and M treatment is the best among the tested amendments with the highest rice grain yield. M treatment increased the OP_{ss} values significantly to relieve the inhibition of the water uptake by plants. Additionally, M treatment significantly enhanced K^+ content and impeded Na^+ accumulation in shoots. SA values were reduced while ST values were increased for all tests with amendments applied. In conclusion, mixture of desulfurization gypsum, sandy soil and farm manure is a better ameliorant for the improvement of rice growth and yield in the western Songnen Plain.

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21 **Abstract**

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32 **Results.** The results indicated that amendments application have positive effects on rice yield, and
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35 Additionally, M treatment significantly enhanced K^+ content and impeded Na^+ accumulation in
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37 applied. In conclusion, mixture of desulfurization gypsum, sandy soil and farm manure is a better
38 ameliorant for the improvement of rice growth and yield in the western Songnen Plain.

39 Introduction

40 Soil salinity-sodicity is one of the central impediments with serious influences on crop productivity
41 and sustainability in arid and semiarid areas (Suarez 2001, Qadir et al. 2006). Saline-sodic soils in China
42 reached approximately 3.67×10^7 ha, and Songnen Plain is one of the major saline-sodic areas (Yao et al.
43 2008, Yang et al. 2016). pH stress and Na^+ toxicity are the main causes of the degradation in saline-sodic
44 soils (Gharaibeh et al. 2010). Efforts have been made to ameliorate saline-sodic soils including the use of
45 chemical amendments, farmyard manure, sandy soil and engineering (Qadir et al. 2007, Ahmad et al. 2013).
46 Chemical amendments usually used as a Ca^{2+} source to replace the exchangeable Na^+ (Oster, 1982). Manure
47 application improves soil structure and alleviates soil sodicity (Yu et al. 2010). Sandy soil amendment
48 changes soil compactness and reduces salt content (Wang et al. 2010).

49 Rice (*Oryza sativa* L.) was planted in order to effectively utilize saline-alkaline soil, due to the fact
50 that irrigation is beneficial to rice growth and salt leaching. Crops growth responds to salinity and sodicity
51 in two phases: a continuous osmotic phase that the potential energy of saline-sodic soil solution was lowered
52 by its osmotic pressure, which inhibits the plants' water uptake; and a slower ionic phase that ion toxicity
53 or ion imbalance due to the plants' accumulation of ions over a period of time (Munns and Tester., 2008).
54 Most amendment study have focused on the characteristics of soil physiochemical properties (Chi et al.
55 2012; Zhao et al. 2018) rather than on the variety of osmotic potential in the soil solution and selective
56 absorption of ions in plant, although they have important effects on crop biomass (Wang et al. 2009).

57 Rice showed moderate sensitivity to salinity and sodicity from the study of Mass et al. (1977). Kelly
58 and Rengasamy (2006) showed that osmotic stress is one of the major factor in reducing crop yield. Tiessen

59 and Carolus (1963) concluded that decreasing the osmotic potential of soil solution below -1.3 bars by
60 applying fertilizer was detrimental to the elongation of roots of plant. Song et al. (1996) demonstrated that
61 the survival of rice plant under saline-sodic conditions is correlated with Na^+ and K^+ accumulations in plant
62 tissues. Yamanouchi et al. (1987) investigated that Na^+ concentrations in shoots are inversely correlated
63 with the relative plant growth and yield. Matsushita et al (1991) found that the susceptibility of rice plants
64 to saline-alkali stress is due to the limited ability to restrict Na^+ transportation to shoots. K^+ is an important
65 macronutrient for the growth of plants and cannot be substituted by Na^+ (Bhandal et al. 1988). The ability
66 of plant to keep a high cytoplasmic K^+/Na^+ ratio is one of the most momentous mechanisms of salt tolerance
67 (Maathuis et al. 1999). 

68 In this study, we analyzed the effect of amendments on osmotic potential in the soil solution,
69 characterized K^+ and Na^+ absorption of rice, and measured several parameters such as K^+ and Na^+ contents
70 in shoots and roots, selective absorption/transport for K^+ over Na^+ , distribution of K^+ , Na^+ in rice organs,
71 and yield of rice under various amendments application.

72 **Materials & Methods**

73 **Location description**

74 The study was conducted from 2009 to 2017 at Da'an Sodic Land Experiment Station (45°35'58"-
75 45°36'28"N, 123°50'27"-123°51'31"E, 132.1 m.a.s.l), operated by Chinese Academy of Sciences.
76 According to USDA texture classification system, the soil at the study site has a clay loam texture.

77 **Field design and treatment**

78 The experiment was arranged in a random block design with three replicates of 20 m² for each plot.
79 There were five treatments implemented: (1) CK, without amendment application; (2) DG, amended with
80 desulfurization gypsum of 3 kg m⁻²; (3) SS, amended with sandy soil of 10 cm thickness; (4) FM, amended
81 with 6 kg m⁻² farmyard manure (5) M, amended with the mixture of desulfurization gypsum, sandy soil and
82 farm manure, the amounts of which are equal to those in the DG, SS and FM treatments. Plastic cloth buried
83 1 m deep in soil is separated from each other between plots to prevent disturbance of lateral movement of
84 amendments, water and salt. Soil amendments were manually mixed with the soil down to 20 cm prior to
85 the start of this experiment in 2009. The local rice cultivar (G19) was planted in this experiment.

86 **Measurements**

87 Rice samples were collected from each plot on October 1, 2017 before harvest and transported to the
88 laboratory. Rice plants were separated into roots, leaves, sheaths and panicles. Plant samples were dried for
89 48h at 80°C in an air-forced oven. Dried materials were finely grounded using a ball mill. They were then
90 digested using an acid mixture [sulphuric acid/ perchloric acid (H₂SO₄/HClO₄=4:1)]. K⁺ and Na⁺
91 concentrations were determined using an atomic absorption spectrometer (GGX-900). K⁺ and Na⁺ contents
92 in the shoot were calculated from K⁺ and Na⁺ contents and dry weights of grains, leaves and sheaths; K⁺
93 and Na⁺ contents in the whole plant were calculated from K⁺ and Na⁺ contents and dry weights of grains,
94 leaves, sheaths and roots.

95 Plants were harvested in October 20, 2017. The following growth and yield were determined and
96 calculated: plant height, panicle length, number of panicles per pot, number of grains per panicle, 1000-
97 grain weight and grain yield (Zeng et al. 2000).

98 Soil samples were obtained from each plot at six depths of 0-10 cm, 10-20 cm, 20-40 cm, 40-60 cm,
99 60-80 cm and 80-100 cm after harvest. All the soil samples were dried at 105°C for 24 h and passed through
100 a 2-mm diameter sieve. The EC values and concentrations of K⁺ and Na⁺ were measured in a soil: water
101 suspension (1: 5) as described by Sumner (1993). The osmotic potential in the soil solution (OP_{ss}) was
102 calculated as follows:

$$103 \quad OP_{ss} = (-0.36) \times 10EC \quad (\text{Bohn et al., 1979})$$

104 **SA and ST Calculation**

105 Selective absorption (SA) and selective transport (ST) values were calculated according to the
106 following formula of Wang (2002) and Wang et al. (2004) using data obtained from the experiments
107 described earlier:

$$108 \quad SA = (\text{K/Na in root dry weight}) / (\text{soil K/Na at 0-40 cm depth})$$

$$109 \quad ST = (\text{K/Na in shoot dry weight}) / (\text{K/Na in root dry weight})$$

110 **Statistical analysis**

111 The obtained results were statistically evaluated by the analysis of variance (ANOVA) method. The
112 differences between mean values were evaluated by the Duncan test. All analyses were done with the

113 SPSS20.0 software (New York, USA), at the level of significance $P=0.05$.

114 **Results**

115 **Soil physicochemical properties**

116 The soil prior to the start of the experiment represents a typically severe saline-sodic soil with pH (1:
117 5 H₂O) at 10.47, electrical conductivity (EC) (1: 5 H₂O) of 2.36 mS cm⁻¹, soil organic carbon (SOC) at 2.80
118 g kg⁻¹ and exchangeable sodium percentage at 79.66% in the top 20-cm soil layer, which is considered to
119 be the effective rooting zone.

120 **Effect of amendments application on OP_{ss}**

121 The osmotic potential can serve as a good index for evaluating plant response to saline-alkali stress
122 (Souza et al., 2012). The osmotic potential in the soil solution (OP_{ss}) was increased by amendments
123 application compared to the control, and in 0-40 cm soil layer, amendments application generally increased
124 the OP_{ss} values in the following order: M>DG>SS>FM>CK (Figure 1). In the same soil layer, the highest
125 OP_{ss} was observed for M, which means that the ability to reduce the salt concentration of soil solution is
126 stronger, followed by DG.

127 **Effect of amendments application on Na⁺, K⁺ contents in rice shoots and roots**

128 The Na⁺ concentration in shoots of rice plants varied with different amendments applied in the saline-
129 sodic soil (Figure 2A). Rice shoots of plants treated with M showed the lowest Na⁺ content of 0.91 mg/g
130 dry weight, compared to Na⁺ content of 0.95, 0.98, 0.99 and 1.01 mg/g dry weight with FM, CK, SS and
131 DG treatments, respectively. The difference on Na⁺ content between DG and M treatments was significant.
132 The mean root Na⁺ content decreased from the maximum 2.38 mg/g dry weight in the CK treatment to 2.36,
133 2.21, 2.16 and 2.02 mg/g dry weight with M, SS, FM and DG treatments, respectively. The differences
134 among amendments treatment and CK were ~~less obvious~~ (Figure 2B). Amendments application
135 significantly enhanced K⁺ content in rice shoots compared to the control treatment, with the highest K⁺
136 content found for DG (Figure 2C). The K⁺ contents in rice roots with M, SS, FM treatments were lower
137 than that with the control treatment. The lowest K⁺ content was observed for FM, which was 16.75% lower
138 than that of CK (Figure 2D).

139 **Selective absorption and transport for K⁺ over Na⁺ in rice plant**

140 Selective absorption (SA) represents the net capacity of a plant to absorb K^+ relative to Na^+ from the
141 shallow soil (0-40 cm); Selective transport (ST) reflects the net capacity of a plant to transport in favor of
142 K^+ over Na^+ from root to shoot (Wang et al. 2004). Compared with the CK, SA values decreased after
143 applying amendments, and the lowest SA value was observed for M, which was 74.8% lower than that of
144 CK (Figure 3A). However, ST values were higher with amendment applications than the one without. The
145 peak ST value among different treatments was found for M, which was 1.5 times more than the ST value
146 of CK (Figure 3B).

147 The mean soil Na^+ content decreased from the maximum (6.68 mmol/L) in the control treatment to
148 3.16, 4.35, 5.11 and 6.60 mmol/L with M, DG, SS and FM treatments, respectively. The differences among
149 M, DG and CK were significant (Figure 4A). Amendment application slightly enhanced K^+ content in the
150 soil compared to CK (Figure 4B). The Ca^{2+} contents of the soil were higher for treatments with amendments
151 than the one without, and differences among the 4 different amendments were not significant (Figure 4C).

152 Amendment application hindered the uptake of K^+ over Na^+ from soil to root compared with CK
153 (Figure 3A), which is probably a consequence of rice physiological adjustment. Amendment application
154 enhanced the uptake of K^+ over Na^+ from root to shoot compared with CK (Figure 3B), which was mainly
155 caused by the strong capacity of selective transport of K^+ over Na^+ under amendments application.

156 **Characteristics of distribution of Na^+ and K^+ in rice with different amendments** 157 **application**

158 There were little differences on Na^+ content in the whole rice plants among different treatments in
159 saline-sodic soils (Figure 5A). K^+ content in the whole plant was significantly enhanced after amendment
160 application, but the differences between the 4 treatments with amendments were limited (Figure 5B). Na^+
161 absorbed by the whole plant was almost the same with and without amendments, which was different from
162 the observations on rice organs (Figure 5A, Table 1). The Na^+ contents in rice roots and grains both
163 decreased when applying amendments in saline-sodic soils; which was contrary to the rise of K^+ contents
164 in sheaths and leaves (Table 1). Compared to the control treatment, DG, SS, FM and M treatments increased
165 the K^+ contents in rice sheaths by 57.2%, 54.9%, 44.1% and 25.5%, respectively.

166 For the distribution of ions in rice organs, there was a higher K^+ proportion in leaves; the distribution

167 of Na^+ was more focused in roots. The ~~fundamental~~ order of accumulation of Na^+ in various organs was
168 roots>leaves>sheaths>grains (Table 1). The order is imposed by the fact that root system retains more Na^+
169 and prevents Na^+ from being transported to the organs aboveground in saline-sodic soils, resulting in higher
170 K^+ proportion in leaves, sheaths and grains, which was illustrated beneficial to normal metabolic (Borsani
171 2001, Ahmad et al., 2005).

172 **Relationship between OP_{ss} , selective absorption and yield of rice**

173 The changes of rice growth and yield were analyzed and the results are summarized in Table 2. The
174 grain yield of rice increased from 4426.01 kg/hm² in CK treatment to 8924.87, 8359.39, 8184.42 and
175 7259.90 kg/hm² in M, DG, SS and FM treatments, respectively. Amendment treatments significantly
176 enhanced the grain yield of rice, however, the differences among different amendments were not significant
177 (Table 2). Soil amendment application generally increased the 1000-grain weight in the following order:
178 M>DG>SS>FM>CK (Table 2). Additionally, M and DG treatments significantly increased the 1000-grain
179 weight to 1.16 and 1.13 times more than CK treatment does, respectively (Table 2). Compared to CK
180 treatment, M treatment significantly increased the mean number of panicles per pot, while FM treatment
181 considerably enhanced the number of grains per panicle (Table 2). There was no obvious difference on rice
182 height and panicle length between various treatments (Table 2).

183 Significant positive relationships were found between $\text{OP}_{0-20\text{cm}}$ and 1000-grain weight ($R^2=0.992$,
184 $P=0.001$, Table 3) and between $\text{OP}_{0-20\text{cm}}$ and grain yield ($R^2=0.946$, $P=0.015$, Table 3). Significant negative
185 relationships were found between SA and rice grain yield ($R^2=0.921$, $P=0.026$, Table 3) and between SA
186 and 1000-grain weight ($R^2=0.884$, $P=0.047$, Table 3) for rice. Furthermore, there was a positive correlation
187 between 1000-grain weight and grain yield ($R^2=0.977$, $P=0.004$, Table 3). There was no significant
188 relationship between either SA or ST and other growth and yield of rice (Table 3).

189 **Discussion**

190 **Characteristics of Na^+ and K^+ absorption in rice**

191 Applying amendments reduces the uptake and accumulation of Na^+ and increases the uptake of K^+
192 (Sudhir et al. 2004), which is consistent with our experimental results. Amendments addition reduce plant
193 Na^+ uptake under saline-sodic soils by transient Na^+ binding due to its high absorption capacity and by

194 releasing mineral nutrients into the soil (Akhtar et al. 2015, Melas et al. 2017, Jin et al. 2018). In addition,
195 as the amendments applied, osmotic stress and Na^+ toxicity were significantly decreased for better plant
196 growth in saline-sodic soils (Swarup 1988, Yuncai et al. 2005, Luo et al. 2018, Shi et al. 2019). Similar to
197 our results, previous studies showed that plants accumulate excessive Na^+ in their shoots under stress caused
198 by high salinity-sodicity (Roy et al. 2014), and the Na^+ content in shoots increased significantly with the
199 surge in soil salinity-sodicity (Syed et al. 2017).

200 Adding amendments reduced the salinity-sodicity stress of the soil (Changanti et al. 2015), therefore,
201 the rice planted in the CK plot was under a higher external salinity-sodicity stress. As a result, SA value of
202 rice plants with CK was higher than those with amendments application to maintain a high cytosolic K^+/Na^+
203 ratio, which is thought to be one of the most important mechanisms of salt tolerance exhibited by plants
204 (Gorham et al. 1990, Dubcovsky et al. 1996, Munns et al. 2008, Munns et al. 2010).

205 **Effects of Ca^{2+} on SA and ST values**

206 Application of amendments enhanced the uptake of K^+ over Na^+ from root to shoot, which was mainly
207 caused by the strong capacity of selective transport of K^+ over Na^+ under amendments. Furthermore, the
208 redundant Na^+ were intercepted by rice roots, this is consistent with previous studies by Mamo et al. (1996)
209 and Abdullah et al. (2001). Amendments consisted of Ca^{2+} promoted K^+ rather than Na^+ absorption,
210 resulting in the enhancement on selectivity of K^+ over Na^+ (Shinsuke et al. 2011). Ca^{2+} can replace Na^+ in
211 plants, which restored cell wall stability and plasma membrane integrity and facilitated K^+/Na^+ selectivity
212 (Zhang et al. 2010, Wu et al. 2012). Ca^{2+} from amendments can also reduce the bypass flow of rice under
213 salinity stress (Anil et al. 2005).

214 It can be concluded in this study that amendments application in saline-sodic soils can reduce K^+
215 selective absorption over Na^+ from soil to root and enhance K^+ selective absorption over Na^+ from root to
216 shoot. However, it was shown that, under saline-sodic conditions, amendments consisted of Ca^{2+}
217 remarkably increased K^+/Na^+ selectivity of both roots and shoots in *Medicago sativa* (Al-Khateeb 2006)
218 and *Cornus sericea* (Renault et al. 2009). In contrast with what Wang et al (2007) found that applications
219 of amendments consisted of Ca^{2+} had no influence on K^+/Na^+ selective absorption. Previous study on rice
220 plants (Wu et al. 2012) indicated that amendment-treated (consisted of Ca^{2+}) samples showed stronger

221 selective absorption for K^+ than Na^+ at low salinity. Besides the difference on amendments used in those
222 studies, the degree of salinity stress was quite unlike from one case to the other.

223 Yield of rice

224 our experiments have shown that application of calcium can enhance the OP_{ss} values, and then decrease
225 osmotic pressure of soil solution, ultimately increase the plant growth and yield of rice in saline-sodic soils.
226 Transient salinity affects the plants' absorption of available water, which result in the reduction in plant
227 yield (Rengasamy. 2010a, 2010b). In saline-sodic soils, application of ameliorants alleviates the salinity-
228 sodicity stress on plants (Irshad et al., 2002). Applying a small amount of calcium could enhance the plants'
229 salt tolerance (Cramer 1992).

230 The grain yield of rice was higher with amendment application compared to that with CK treatment.
231 The negative relationship between grain yield and SA values indicates that high SA value from the
232 promoted plant growth caused by Na^+ uptake is of little benefit to rice plants (Milford et al. 1977, Durrant
233 et al. 1978). A possible explanation for this result is that the low SA values of plant was partly ascribed to
234 its relative sensitivity to root which increased the Na^+ accumulation in root and therefore decreased the SA
235 values (Leland et al. 1999, Ren et al. 2005, Tsialtas et al 2009).

236 Amendments are known to improve root environment and increase rice yield (Abrishamkesh et al.
237 2015). In this study, we found that amendment application can reduce the absorption of Na^+ in rice shoots,
238 which could improve rice yield. However, the mechanism of physiological and ecological changes in rice
239 plants, such as enzymes, photosynthetic absorption, membrane permeability and etc, has not been well
240 investigated, especially for the saline-sodic soils. The work present here is of great worth to better
241 understandings of how rice plants behave in saline-sodic soils through further exploitations.

242 Conclusions

243 In this long-term field experiment, the amendments application significantly increased the yield of
244 rice. Relative to CK treatment, the FM and M treatments significantly enhanced the 1000-grain weight and
245 the SS and FM treatments significantly improved the number of grains per panicle and the number of
246 panicles per pot, respectively. Furthermore, all amendments application significantly increased the grain
247 yield of rice. In particularly, the M treatment is the best among the tested amendment treatments with the

248 highest rice grain yield in the saline-sodic soils. Because the M treatment increased the OP_{ss} significantly,
249 and then, it can relieve the inhibition of the water uptake by plants. In addition, a positive effect of
250 amendments application on reducing Na^+ accumulation and increasing the uptake of K^+ of rice shoot, and
251 amendments application can increase ST values and decrease SA values. Moreover, there exist an ion
252 regionalization distribution in rice plant: a higher K^+ proportion in leaves and a higher Na^+ proportion in
253 roots. Collectively, mixture of desulfurization gypsum, sandy soil and farm manure is a better selection for
254 increasing the yield of rice in saline-alkaline soils in the western Songnen Plain.

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257 References

- 258 Abdullah Z, Khan MA, Flowers TJ. 2001. Causes of sterility and seed set in rice under salinity stress. *Journal of*
259 *Agronomy and Crop Science* **187**: 25-32.
- 260 Abrishamkesh S, Gorji M, Asadi H. 2015. Effects of rice husk biochar application on the properties of alkaline soil
261 and lentil growth. *Plant, Soil and Environment* **61**: 475-482.
- 262 Ahmad R, Jabeen R. 2005. Foliar spray of mineral elements antagonistic to sodium a technique to induce salt tolerance
263 in plants growing under saline conditions. *Pak. J. Bot* **37**: 913-920.
- 264 Ahmad S, Ghafoor A, Akhtar ME, Khan MZ. 2013. Ionic displacement and reclamation of saline-sodic soils using
265 chemical amendments and crop rotation. *Land Degradation & Development*. **24**: 170-178.
- 266 Akhtar SS, Andersen MN, Liu F. 2015. Residual effects of biochar on improving growth, physiology and yield of
267 wheat under salt stress. *Agricultural Water Management* **158**: 61-68.
- 268 Al-khateeb SA. 2006. Effect of calcium/sodium ratio on growth and ion relations of alfalfa (*Medicago sativa* L.)
269 seedling s growth under saline condition. *Journal of Agronomy* **5**: 175-181.
- 270 Anil VS, Kirishnamurthy P, Kuruvilla S, Suchartha K, Thomas G, Mathew MK. 2005. Regulation of the uptake and
271 distribution of Na^+ in shoots of rice (*Oryza sativa*) variety Pokkali: role of Ca^{2+} in salt tolerance response.
272 *Physiologia Plantarum* **124**: 451-464.
- 273 Bhandal IS, Malik CP. 1988. Potassium estimation, uptake, and its role in the physiology and metabolism of flowering
274 plants. *International Review of Cytology* **110**: 205-254.

- 275 Bohn HL, Myer RA, O'Connor GA. 2002. Soil chemistry. John Wiley & Sons.
- 276 Borsani O. 2001. Identification of two loci in tomato reveals distinct mechanisms for salt tolerance. *The Plant Cell*
- 277 *Online* **13**: 873-888.
- 278 Chaganti VN, Crohn DM. 2015. Evaluating the relative contribution of physiochemical and biological factors in
- 279 ameliorating a saline-sodic soil amended with composts and biochar and leached with reclaimed water. *Geoderma*
- 280 **259-260**: 45-55.
- 281 Chi CM, Zhao CW, Sun XJ, Wang ZC. 2012. Reclamation of saline-sodic soil properties and improvement of rice
- 282 (*Oriza sativa* L.) growth and yield using desulfurized gypsum in the west of Songnen Plain, northeast China.
- 283 *Geoderma* **187**: 24-30.
- 284 Corham J, Jones RGW, Bristol A. 1990. Partial characterization of the trait for enhanced K^+ - Na^+ discrimination in the
- 285 Dgenome of wheat. *Planta* **180**: 590-597.
- 286 Cramer GR. 1992. Kinetics of maize leaf elongation. II. Responses of a Na-excluding cultivar and a Na-including
- 287 cultivar to varying Na/Ca salinities. *Journal of Experimental Botany* **43**: 857-864.
- 288 Dubcovsky J, Santa MG, Epatin E, Luo MC, Dvorak J. 1996. Mapping of the K^+ / Na^+ discrimination locus *Knal* in
- 289 wheat. *Theoretical and Applied Genetics* **92**: 448-454.
- 290 Durrant MJ, Draycott AP, Milford GFJ. 1978. Effect of sodium fertilizer on water status and yield of sugar beet.
- 291 *Annals of applied biology* **88**: 321-328.
- 292 Gharaibeh MA, Eltaif NI, Shra'Ah SH. 2010. Reclamation of a calcareous saline-sodic soil using phosphoric acid and
- 293 by-product gypsum. *Soil Use and Management* **26**: 141-148.
- 294 Giovanna. 2017. Can Biochar Protect Labile Organic Matter Against Mineralization in Soil?. *Pedosphere* **27**: 822-
- 295 831.
- 296 Irshad M, Honna T, Eneji AE, Yamamoto S. 2002. Wheat response to nitrogen source under saline conditions. *Journal*
- 297 *of Plant Nutrition* **25**: 2603-2612.
- 298 Jin F, Ran C, Anwari QA., Geng YQ., Guo LY, Li JB., Han D, Zhang XQ, Liu X, Shao XW. 2018. Effects of biochar
- 299 on sodium ion accumulation, yield and quality of rice in saline-sodic soil of the west of Songnen plain, northeast
- 300 China. *Plant Soil Environment* **64**: 612-618.
- 301 Kelly J, Rengasamy P. 2006. Diagnosis and management of soil constraints: transient salinity, sodicity and alkalinity.

- 302 The University of Adelaide and Grain Research and Development Corporation, Australia.
- 303 Leland E, Eugene V. 1999. Crop response and management of salt-affected soils. Handbook of plant and crop stress.
- 304 169.
- 305 Luo SS, Wang SJ, Lei T, Shi SH, Xu SQ., Yang F., Li XJ., Wang ZC., Tian CJ. 2018. Aggregate-related changes in
- 306 soil microbial communities under different ameliorant applications in saline-sodic soils. *Geoderma* **329**: 108-117.
- 307 Maas EV, Hoffman GJ. 1977. Crop salt tolerance—current assessment. *Journal of the irrigation and drainage division*
- 308 **103**: 115-134.
- 309 Maathuis FJ, Amtmann A. 1999. K⁺ nutrition and Na⁺ toxicity: the basis of cellular K⁺/Na⁺ ratios. *Annals of Botany*
- 310 **84**: 123–133.
- 311 Mamo T, Richter C, Heiligttag B. 1996. Salinity effects on the growth and ion contents of some chickpea (*Cicer*
- 312 *arietinum*) and lentil (*Lens culinaris medic*). *Journal of Agricultural Science* **113**: 407-410.
- 313 Matsushita N, Matoh T. 1991. Characterization of Na⁺ exclusion mechanisms of salt-tolerant reed plants in
- 314 comparison with salt-sensitive rice plants. *Plzysiol. Plam* **83**: 170-176.
- 315 Milford GFJ, Cormack WF, Durrant MJ. 1977. Effects of sodium chloride on water status and growth of sugar beet.
- 316 *Journal of Experimental Botany* **28**: 1380-1388.
- 317 Mori S, Suzuki K, Oda R, Higuchi K, Maeda Y, Yoshiba M, Tadano T. 2011. Characteristics of Na⁺ and K⁺ absorption
- 318 in *Suaeda salsa* (L.) Pall. *Soil Science and Plant nutrition* **57**: 377-386.
- 319 ~~Munns R, James RA. 2008. Mechanisms of salinity tolerance. *Annual Review of Plant Biology* **59**: 651–681.~~
- 320 Munns R, Tester M. 2008. Mechanism of salinity of salt tolerance. *Annual Review of Plant Biology* **59**: 651-681.
- 321 Munns R, Wallace PA, Teakle NL, Colmer TD. 2010. Measuring soluble ion concentrations (Na⁺, K⁺, Cl⁻) in salt-
- 322 treated plants. In plant Stress Tolerance, Methods and Protocols. New York, USA, Human press, Springer Science.
- 323 Oster JD. 1982. Gypsum usage in irrigated agriculture: a review. *Fertilizer Research* **3**: 73-89.
- 324 Qadir M, Noble A, Schubert S. 2006. Sodicty-induced land degradation and its sustainable management: Problems
- 325 and prospects. *Land Degradation & Development* **17**: 661-676.
- 326 Qadir M, Schubert S, Badia D, Sharma BR, Qureshi AS, Murtaza G. 2007. Amelioration and nutrient management
- 327 strategies for sodic and alkali soils. *CAB Rev. Perspect Agric. Vet. Sci. Nutr. Nat. Resour* **21**: 1-13.
- 328 Ren ZH, Gao JP, Li LG. 2005. A rice quantitative trait locus for salt tolerance encodes a sodium transporter. *Nature*

- 329 *genetics* **37**: 1141.
- 330 Renault S, Affifi M. 2009. Improving NaCl resistance of red-osier dogwood: role of CaCl₂ and CaSO₄. *Plant and soil*
- 331 **315**: 123-133.
- 332 Rengasamy P. 2010a. Osmotic and ionic effects of various electrolytes on the growth of wheat. *Australian Journal of*
- 333 *Soil Research* **48**: 120-124.
- 334 Rengasamy P. 2010b. Soil processes affecting crop production in salt-affected soils. *Functional Plant Biology* **37**:
- 335 613-620.
- 336 Roy C, Mishra R. 2014. Impact of NaCl stress on the physiology of four cultivars of *S. lycopersicum*. *Res. Plant Biol*
- 337 **4**: 09-20.
- 338 Shi SH, Tian L, Nasir F, Bahadur A, Batool A, Luo SS, Yang F, Wang ZC, Tian CJ. 2019. Response of microbial
- 339 communities and enzyme activities to amendments in saline-alkaline soils. *Applied Soil Ecology* **135**: 16-24.
- 340 Song JQ, Fujiyama H. 1996. Difference in response of rice and tomato subjected to sodium salinization to the addition
- 341 of calcium. *Soil Science and Plant Nutrition* **42**: 503-510.
- 342 Souza ERD, Freire MBGDS, Karina Patricia Vieira da Cunha. 2012. Biomass, anatomical changes and osmotic
- 343 potential in *Atriplex nummularia* Lindl. cultivated in sodic saline soil under water stress. *Environmental &*
- 344 *Experimental Botany* **82**: 20-27.
- 345 Suarez DL. 2001. Sodic soil reclamation: Modelling and field study. *Soil Research* **39**: 1225-1246.
- 346 Sumner ME. 1993. Sodic soils: New perspectives. *Australian Journal of Soil Research* **31**: 683-750.
- 347 Sun H, Lu H, Chu L, Shao H, Shi W. 2017. Biochar applied with appropriate rates can reduce N leaching, keep N
- 348 retention and not increase NH₃ volatilization in a coastal saline soil. *Science of the Total Environment* **575**: 820-
- 349 825.
- 350 Sushir P, Murthy SDS. 2004. Effects of salt stress on basic processes of photosynthesis. *Photosynthetica* **42**: 481-486.
- 351 Swarup A. 1982. Availability of ions, zinc and phosphorus in submerged sodic soil as affected by amendments during
- 352 the growth period of rice crop. *Plant and Soil* **66**: 37-43.
- 353 Syed GA, Abdur R. 2017. The influence of salinity and drought stress on sodium, potassium and proline content of
- 354 *Solanum lycopersicum* L. cv. Rio granded. *Pak. J. Bot.* **49**: 1-9.
- 355 Tiessen H, Carolus RL. 1963. Effect of different analyses and concentrations of fertilizer solutions on initial root

- 356 growth of tomato and tobacco plants. *Proc. Am. Soc. Hort. Sci* **83**: 680-683.
- 357 Tsialtas JT, Maslaris N. 2009. Selective absorption of K over Na in sugar beet cultivars and its relationship with yield
358 and quality in two contrasting environments of central Greece. *Journal of agronomy and crop science* **195**: 384-
359 392.
- 360 Wang CM, Zhang JL, Liu XS, Li Z, Wu GQ, Cai JY. 2009. *Puccinellia tenuiflora* maintains a low Na⁺ level under
361 salinity by limiting unidirectional Na⁺ influx resulting in a high selectivity for K⁺ over Na⁺. *Plant, Cell and*
362 *Environment* **32**, 486-496.
- 363 Wang S, Wan C, Wang Y, Chen H, Zhou Z, Fu Y, Sosebee RE. 2004. The characteristics of Na⁺, K⁺ and free proline
364 distribution in several drought resistant plants of the Alxa Desert, China. *J. Arid Environ* **56**: 525-539.
- 365 Wang S, Zheng W, Ren J, Zhang C. 2002. Selectivity of various types of salt-resistant plants for K⁺ over Na⁺. *J. Arid*
366 *Environ* **52**: 457-472.
- 367 Wang SM, Zhang JL, Flower TJ. 2007. Low-affinity Na⁺ uptake in the halophate *Suaeda maritima*. *Plant Physiology*
368 **145**: 559-571.
- 369 Wang SM, Zhao GO, Gao YS, Tang ZC, Zhang CL. 2004. *Puccinellia tenuiflora* exhibits stronger selectivity for K⁺
370 over Na⁺ than wheat. *Journal of Plant Nutrition* **27**: 1841-1857.
- 371 Wang Y, Yang C, Liu G. 2007. Development of a cDNA microarray to identify gene expression of *Puccinellia*
372 *tenuiflora* under saline-alkali stress. *Plant Physiology and Biochemistry* **45**:567-576.
- 373 Wang MM, Liang ZW, Wang ZC, Huang LH, Ma HY, Liu M, Gu X. 2010. Effect of sand application and flushing
374 during the sensitive stages on rice biomass allocation and yield in a saline-sodic soil. *J. Food Agric. Environ* **8**: 692-
375 697.
- 376 Wu GQ, Wang SM. 2012. Calcium regulates K⁺/Na⁺ homeostasis in rice (*Oryza sativa* L.) under saline conditions.
377 *Plan Soil and Environment* **58**: 121-127.
- 378 Yamanouchi M, Maeda Y, Nagai T. 1987. Relationship between the salt accumulation in the shoots and the degree of
379 salinity tolerance in rice cultivars. *Soil Sci. Plant Nutr* **58**: 591-594.
- 380 Yang F, An FH, Ma HY, Wang ZC, Zhou X, Liu Z. 2016. Variations on soil salinity and sodicity and its driving
381 factors analysis under microtopography in different hydrological conditions. *Water* **8**: 227.
- 382 Yao RJ. 2008. Development and prospect of the research on salt-affected soils in China. *Acta Pedol. Sin* **45**: 837-845

383 (in Chinese).

384 Yu JB, Wang ZC, Meixner FX, Yang F, Wu HF, Chen XB. 2010. Biogeochemical characterizations and reclamation
385 strategies of saline sodic soil in northeastern China. *Clean-Soil, Air, Water* 38: 1010-1016.

386 Yuncai H, Schmidhalter U. 2005. Drought and salinity: A comparison of the effects of drought and salinity. *J. Plant*
387 *Nutr. Soil Sci* 168: 247-273.

388 Zeng L, Shannon MC. 2000. Salinity Effects on Seedling Growth and Yield Components of Rice. *Crop Science* 40:
389 996.

390 Zhang JL, Flowers TJ, Wang SM. 2010: Mechanisms of sodium uptake by roots of higher plants. *Plant and Soil* 326:
391 45.

392 Zhao YG, Wang SJ, Li Y, Liu J, Zhuo YQ, Chen HX, Wang J, Xu LZ., Sun ZT. 2018. Extensive reclamation of saline-
393 sodic soils with flue gas desulfurization gypsum on the Songnen Plain, Northeast China. *Geoderma*. 321: 52-60.

394

395 **Tables**

396 Table 1. Na⁺, K⁺ contents and K⁺/Na⁺ ratio in different organs of rice plant with various treatments.

397

398 Table 2. Effects of amendments application on growth and yield of rice plant

399

400 Table 3. Correlation coefficients among OP, SA, ST values and different growth and yield of rice..

401

402 **Figures**

403 Figure 1. Osmotic potential of the soil solution with different amendments application. CK,
404 control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM,
405 farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure. Bars
406 represent the standard error of the mean of three replications.

407

408 Figure 2. Na⁺, K⁺ contents in different parts of rice plants with various treatments: CK, control,
409 without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard

410 manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure. Bars represent
411 the standard error of the mean of three replications. Different letters denote means that are
412 significantly different from each other ($P < 0.05$), NS indicates non-significant difference among
413 treatments ($P > 0.05$).

414

415 Figure 3. Selective absorption and selective transport of rice with various treatments: CK, control,
416 without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard
417 manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure. Bars represent
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420 treatments ($P > 0.05$).

421

422 Figure 4. Na^+ , K^+ and Ca^{2+} contents in the soil (0-40 cm) with various treatments: CK, control,
423 without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard
424 manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure. Bars represent
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428

429 Figure 5. Na^+ , K^+ contents in the whole rice plant with various treatments: CK, control, without
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431 mixture of desulfurization gypsum, sandy soil and farmyard manure. Bars represent the standard
432 error of the mean of three replications. Different letters denote means that are significantly
433 different from each other ($P < 0.05$), NS indicates non-significant difference among treatments
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Figure 1(on next page)

Osmotic potential of the soil solution with different amendments application.

CK, control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure.

Bars represent the standard error of the mean of three replications.

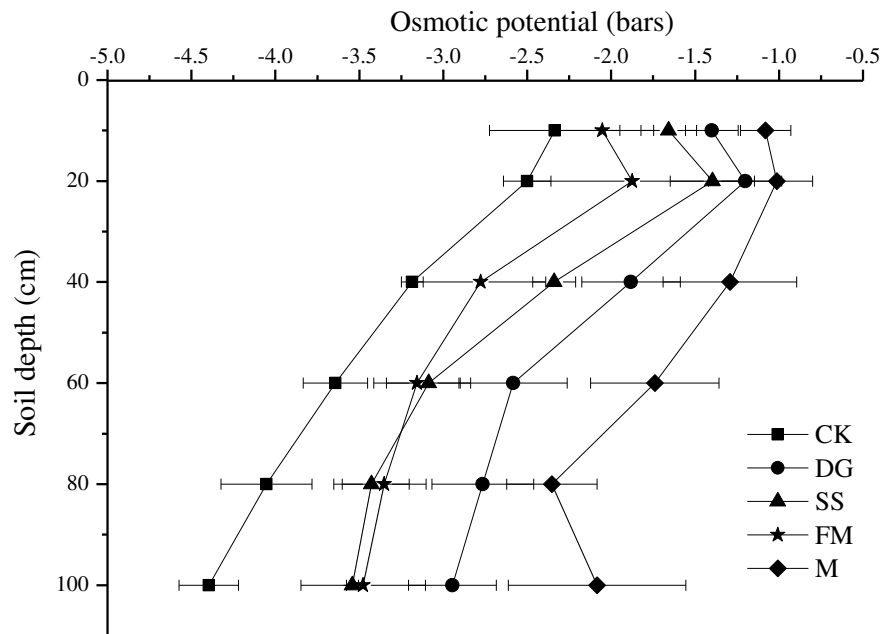


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Figure 2 (on next page)

Na⁺, K⁺ contents in different parts of rice plants with various treatments.

CK, control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure.

Bars represent the standard error of the mean of three replications. Different letters denote means that are significantly different from each other ($P < 0.05$), NS indicates non-significant difference among treatments ($P > 0.05$).

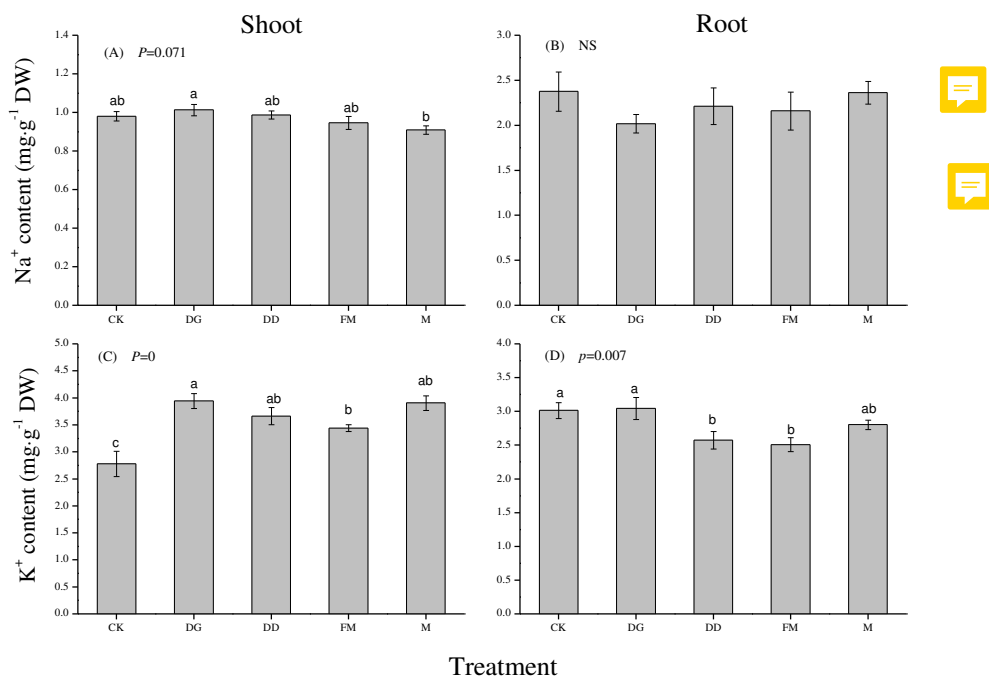


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Figure 3(on next page)

Selective absorption and selective transport of rice with various treatments.

CK, control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure.

Bars represent the standard error of the mean of three replications. Different letters denote means that are significantly different from each other ($P < 0.05$), NS indicates non-significant difference among treatments ($P > 0.05$).

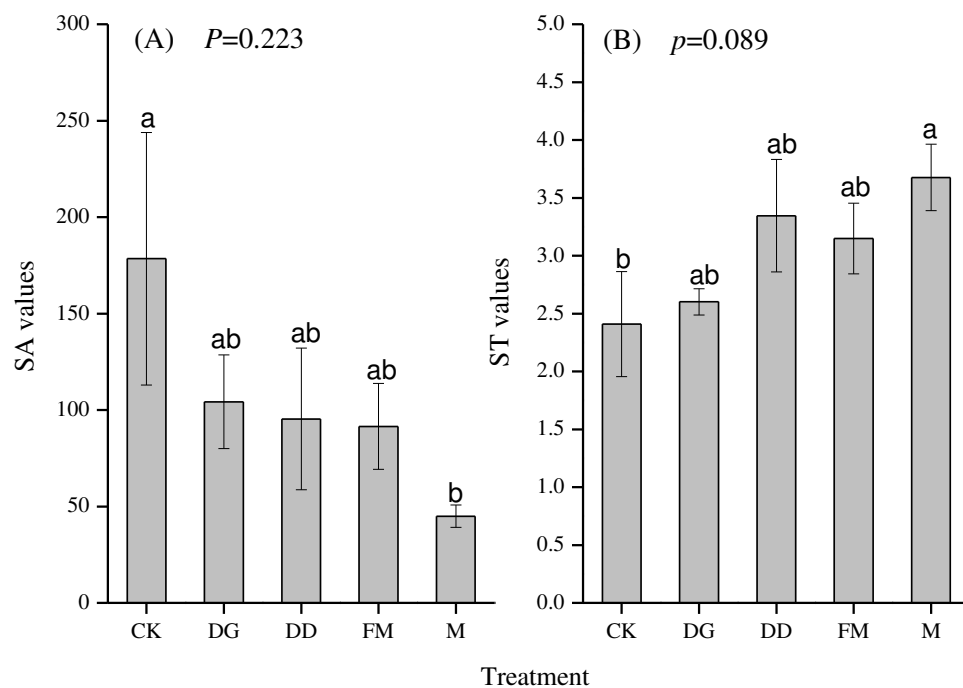


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Figure 4(on next page)

Na⁺, K⁺ and Ca²⁺ contents in the soil (0-40 cm) with various treatments.

CK, control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure.

Bars represent the standard error of the mean of three replications. Different letters denote means that are significantly different from each other ($P < 0.05$), NS indicates non-significant difference among treatments ($P > 0.05$).

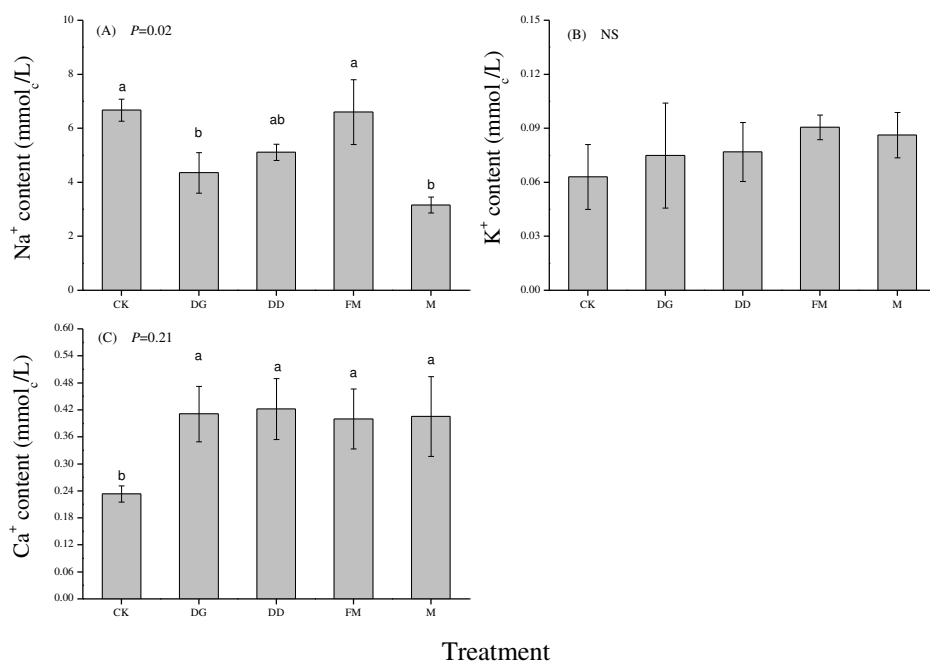


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Figure 5(on next page)

Na⁺, K⁺ contents in the whole rice plant with various treatments.

CK, control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure.

Bars represent the standard error of the mean of three replications. Different letters denote means that are significantly different from each other ($P < 0.05$), NS indicates non-significant difference among treatments ($P > 0.05$).

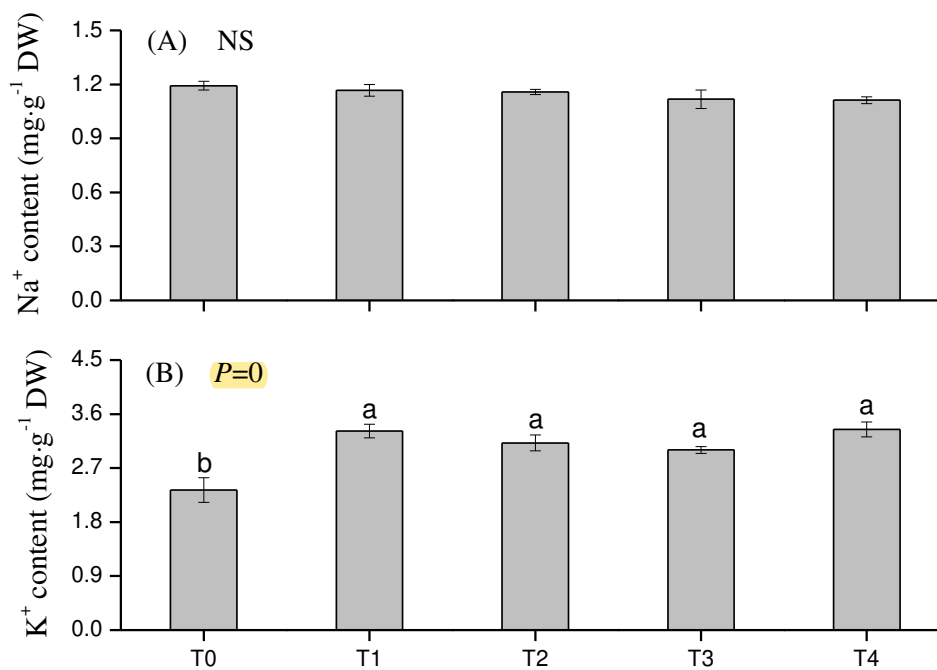


Figure 5. Na⁺, K⁺ contents in the whole rice plant with various treatments: CK, control, without amendments application; DG, desulfurization gypsum; SS, sandy soil; FM, farmyard manure; M, mixture of desulfurization gypsum, sandy soil and farmyard manure. Bars represent the standard error of the mean of three replications. Different letters denote means that are significantly different from each other ($P < 0.05$), NS indicates non-significant difference among treatments ($P > 0.05$).

Table 1 (on next page)

Na⁺, K⁺ contents and K⁺/Na⁺ ratio in different organs of rice plant with various treatments.

The small letters after data indicate that ion contents stand significant difference for $P=0.05$.

1 Table 1. Na⁺, K⁺ contents and K⁺/Na⁺ ratio in different organs of rice plant with various treatments.

Amendment	Organ	Na ⁺ (mg/g DW)	K ⁺ (mg/g DW)	K ⁺ /Na ⁺
CK	Grain	0.53c	2.75ab	7.07a
	Leaf	1.38b	3.61a	2.57b
	Sheath	1.06b	1.99b	1.90b
	Root	2.38a	3.01a	1.36b
DG	Grain	0.25d	2.62b	10.61a
	Leaf	1.63b	5.06a	3.18b
	Sheath	1.30c	4.42a	3.40b
	Root	2.02a	3.04b	1.53c
SS	Grain	0.27d	2.69c	10.11a
	Leaf	1.59b	4.93a	3.11b
	Sheath	1.19c	3.56b	2.93b
	Root	2.21a	2.57c	1.28c
FM	Grain	0.19d	2.60b	15.15b
	Leaf	1.61b	5.19a	3.23b
	Sheath	1.13c	2.67b	2.40a
	Root	2.16a	2.51b	1.25b
M	Grain	0.24c	2.61b	12.59a
	Leaf	1.27b	4.62a	3.85b
	Sheath	1.31b	4.65a	3.54b
	Root	2.36a	2.80b	1.21c

2 Note: The small letters after data indicate that ion contents stand significant difference for $P=0.05$.

3

Table 2 (on next page)

Effects of amendments application on growth and yield of rice plant.

mean value and its standard error (SE) are reported. Different letters denote means that are significantly different from each other ($P < 0.05$).

1

Table 2. Effects of amendments application on growth and yield of rice plant

Treatment	Height (cm)	Panicle length (cm)	Number of panicles per pot	Number of grains per panicle	1000-grain weight (g)	Grain yield (kg/ha)
CK	89.00±3.19a	14.25±0.43a	15.75±0.75b	59.25±3.94b	19.36±0.62b	4426.01±684.77b
DG	87.83±3.38a	15.29±0.41a	16.83±0.48b	78.50±9.53ab	21.67±0.56ab	8184.42±1067.62a
SS	89.83±2.06a	15.33±0.59a	16.50±1.26b	86.67±5.16a	20.67±0.79ab	7259.90±508.66a
FM	94.17±1.05a	14.29±0.32a	22.33±1.96a	68.94±8.38ab	22.44±0.71a	8924.874±607.16a
M	92.50±2.60a	14.25±0.25a	18.83±1.14ab	78.17±4.57ab	21.93±0.98a	8359.39±938.64a

2 Note: mean value and its standard error (SE) are reported. Different letters denote means that are significantly

3 different from each other ($P<0.05$)

4

Table 3 (on next page)

Correlation coefficients among OP, SA, ST values and different growth and yield of rice.

* and ** mean significant correlation at 0.05 and 0.01 level, respectively.

1 Table 3. Correlation coefficients among OP, SA, ST values and different growth and yield of rice

	OP _{0-20cm} (bars)	SA value	ST value	Height (cm)	Panicle length (cm)	Number of panicles per pot	Number of grains per panicle	1000-grain weight (g)
SA	-0.857							
ST	0.628	-0.879*						
Height	0.695	-0.589	0.278					
Panicle length (cm)	-0.146	-0.205	0.391	-0.62				
Number of panicles per pot	0.858	-0.772	0.592	0.905*	-0.454			
Number of grains per panicle	0.276	-0.492	0.319	-0.08	0.727	-0.086		
1000-grain weight (g)	0.992**	-0.884*	0.671	0.619	-0.024	0.802	0.375	
Theoretical yield (kg/ha)	0.946*	-0.921*	0.708	0.536	0.161	0.708	0.559	0.977**

2 Note: * and ** mean significant correlation at 0.05 and 0.01 level, respectively.

3