



## Interaction of salinity and cadmium stresses on mineral nutrients, sodium, and cadmium accumulation in four barley genotypes<sup>\*</sup>

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**Abstract:** Interaction of salinity (NaCl) and cadmium (Cd) on growth, mineral nutrients, Na and Cd accumulation in four barley genotypes differing in salt tolerance was studied in a hydroponic experiment. Cd, NaCl and their combined stresses reduced Ca and Mg concentrations in roots and shoots, K concentration in shoots, increased K and Cu concentrations in roots relative to control, but had non-significant effect on micronutrients Cu, Fe and Mn concentrations in shoot. The three stresses reduced accumulation of most tested nutrients in both roots and shoots, except NaCl and NaCl+Cd stresses for root K and shoot Cu accumulation in salt tolerant genotypes. The salt tolerant genotypes did not have higher nutrient concentration and accumulation than the sensitive ones when exposed to Cd and NaCl stresses. In conclusion, the affecting mechanism of Cd stress on nutrients was to some extent different from salinity stress, and the NaCl+Cd stress was not equal to additional Cd and NaCl stresses, probably due to the different valence and competitive site of Na<sup>+</sup> and Cd<sup>2+</sup>. NaCl addition in the Cd-containing medium caused remarkable reductions in both Cd concentration and accumulation, with the extent of reduction being also dependent on genotypes. The salt-tolerant genotypes had lower Na concentration than sensitive ones.

**Key words:** Barley, Cadmium, Growth, Mineral nutrient, Salinity

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### INTRODUCTION

Salinity is one of the most important abiotic stresses limiting crop production. Approximately, one third of the world's irrigated soils and a large proportion of soils in dry-land agricultural regions are saline (Schachtman and Liu, 1999). Salinization is still expanding, posing a threat to sustainable agriculture development. Similarly, cadmium contamination in arable soils and surface water has become severe due to improper management of waste and application of chemicals containing Cd, especially in developing countries (Helal *et al.*, 1999; Mühling and

Läuchli, 2003). In arid and semi-arid regions, it is likely that biosolids, which contain heavy metals such as Cd, are used on saline soils in order to improve soil quality (Weggler-Beaton *et al.*, 2000; Mühling and Läuchli, 2003). Furthermore, soil salinity has been shown to increase Cd concentration in crops grown on soils fertilized with phosphorous fertilizers containing Cd (McLaughlin *et al.*, 1994; Smolders *et al.*, 1997). Thus, interaction of Cd and salinity should be taken into consideration where both stresses are expected to impact crop growth and yield.

In China, there are a huge number of saline and latent saline soils, with total area of about  $9.91 \times 10^7$  ha (Wang *et al.*, 1993). In the past two decades, phosphorous fertilizers were extensively applied to these saline soils to alleviate salt stress to crops (Deng *et al.*, 2002). As a result, Cd content in these soils has been dramatically increased (Lu *et al.*, 1992; He *et al.*,

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1998). Thus, plants grown in salt-affected soil encounter both salinity and Cd stresses.

The inhibition of Cd stress on plant growth is related to its effect on nutrient uptake and distribution. The uptake and translocation of mineral nutrients such as Fe, Zn, Cu and Mn under Cd stress have been reported in crops, such as soybean (Cataldo *et al.*, 1983; Dražić *et al.*, 2004), rice (Rubio *et al.*, 1994; Liu *et al.*, 2003a), wheat (Zhang *et al.*, 2002) and barley (Wu and Zhang, 2002; Wu *et al.*, 2003). Salinity also presents several challenges to plant growth, including nutrient deficiencies and disorders (Neue *et al.*, 1998; Santos *et al.*, 2002). A large number of studies demonstrated that salinity reduced nutrient uptake and accumulation or affected nutrient partitioning within the plant (Liang, 1999; Grattan and Grieve, 1999; Essa, 2002; Smýkalova and Zamecnikova, 2003; Fernández-García *et al.*, 2004), and the differences in ion partitioning and the maintenance of higher nutrients such as  $K^+$  and  $Ca^{2+}$  to  $Na^+$  ratios, especially in young growing and recently expanded tissues, would appear to be important mechanisms contributing to the improved salt tolerance (Wei *et al.*, 2003). It has been well documented that NaCl and Cd stress in combination resulted in more severe growth inhibition of barley plants than Cd or NaCl stress alone (Smýkalova and Zamecnikova, 2003), yet interaction of salinity and cadmium stresses on mineral nutrients is little known. Thus, it is imperative to elucidate whether the interaction of salinity and Cd leads to a further influence on nutrient metabolism for understanding the combinational stress of NaCl and Cd on plants. In the present research, we studied the combined stress of salinity (NaCl) and Cd on mineral nutrients, Na and Cd accumulation in four barley genotypes differing in salt tolerance.

## MATERIALS AND METHODS

### Plant materials and treatments

The experiment was carried out in 2004 at Hua-jiachi campus, Zhejiang University, Hangzhou, China. Four barley (*Hordeum vulgare* L.) genotypes, differing in salt tolerance (tolerant: Gebeina and Zhou 1; sensitive: Newgoutei and Quzhou), evaluated by the previous screening experiment (Huang *et al.*, 2006), were used and seeds of all four genotypes were sur-

face sterilized in 3%  $H_2O_2$  for 20 min, rinsed with distilled water 5 times and germinated in moist quartz sand in a greenhouse. When seedlings grew the second leaf (10 d old), they were selected for uniformity and transplanted onto a 30-L rectangular container, which was covered with a plastic plate with evenly spaced holes and placed in a greenhouse. The composition of the basic nutrient solution was (mg/L):  $(NH_4)_2SO_4$  48.2,  $MgSO_4$  65.9,  $K_2SO_4$  15.9,  $KNO_3$  18.5,  $Ca(NO_3)_2$  59.9,  $KH_2PO_4$  24.8, Fe-citrate 5,  $MnCl_2 \cdot 4H_2O$  0.9,  $ZnSO_4 \cdot 7H_2O$  0.11,  $CuSO_4 \cdot 5H_2O$  0.04,  $HBO_3$  2.9,  $H_2MoO_4$  0.01, and pH in the solution was 6.4. One week after transplanting to the basic nutrient solution, NaCl and  $CdCl_2$  were added to corresponding containers to form the following 4 treatments: (1) control; (2) 2  $\mu mol/L$  Cd; (3) 150 mmol/L NaCl; (4) 2  $\mu mol/L$  Cd+150 mmol/L NaCl. The experiment was laid out as a completely randomized design with 3 replications. The nutrient solution in the containers was continuously aerated with pumps and renewed every 7 d.

### Measurements

Forty-five days after transplanting, plants of each treatment were harvested, roots dipped in 0.1  $\mu mol/L$  EDTA for 30 min, and washed thoroughly with distilled water, and then separated into roots and shoots, dried in an oven at 80 °C and weighed.

The nutrient concentration was determined by an atomic absorption spectrophotometer (Shimadzu, Japan) after the samples were digested in a mixture of  $HNO_3$ - $HClO_4$  (2:1, v/v). Chlorophyll concentration, as expressed by SPAD value was measured on the topmost secondary fully expanded leaf using a chlorophyll meter (Minolta Co. Ltd., Japan).

### Statistical analysis

All data were subjected to two-way ANOVA using the statistical software of SPSS 11.0 for windows and the means among treatments were compared with Duncan's multiple range test.

## RESULTS AND ANALYSIS

### Chlorophyll concentration, plant height and biomass

In comparison with the control, NaCl or Cd

alone and the combination of NaCl and Cd treatments led to significant decline in chlorophyll concentration expressed as SPAD value, plant height and biomass in all genotypes, but no significant difference was found between Cd treatment and control for plant height and root biomass of Quzhou and for shoot biomass of Zhou 1. On the whole, the inhibiting effect of NaCl on the three parameters was more severe than that of Cd (Table 1). Hence, there were significant differences between the two treatments in shoot biomass of all genotypes, in root biomass of all genotypes but Gebeina, and plant height of Quzhou. Moreover, the effect of the combined stress (NaCl+Cd) on the three parameters was basically similar to that of NaCl alone. The difference between the two treatments was only significant for SPAD value of Quzhou and plant height of Gebeina, indicating that 150 mmol/L NaCl produced more severe damage on these examined parameters than 2  $\mu\text{mol/L}$  CdCl<sub>2</sub>.

#### Macronutrient concentration and accumulation

Cd addition increased significantly root K concentration of Gebeina, and decreased shoot K concentration of all four genotypes. Addition of NaCl, on the whole, increased root K concentration in Gebeina and Zhou 1, and reduced shoot K concentration in all four genotypes but Zhou 1 (Table 2). When the plants were exposed to Cd+NaCl treatment, root K concentration increased and shoot K concentration decreased

significantly for all genotypes except for the root K concentration of Newgoutei. The three stresses caused significant decrease in K accumulation in shoots relative to control. But for root K accumulation, Cd treatment did not lead to the reduction, and the effect of both NaCl and NaCl+Cd varied with barley genotypes. Hence, Newgoutei and Quzhou showed significant reduction in the two treatments relative to the control, while Gebeina and Zhou 1 showed obvious increase (Table 3).

Both NaCl and Cd stresses alone decreased Ca concentration in both roots and shoots of all genotypes except for the root Ca concentration of Quzhou and for the shoot Ca concentration of Zhou 1 in Cd treatment, whereas Ca concentration was similar to that of the control (Table 2). Under the combined stress, shoot Ca concentration was significantly lower than that under Cd alone stress, but had no significant difference from that under NaCl stress alone. No significant difference was found in root Ca concentration between combined stress and NaCl stress alone, except that Gebeina had lower root Ca concentration in combined stress than in NaCl stress. There was a marked difference in shoot Ca concentration between salt tolerant and sensitive genotypes in NaCl and NaCl+Cd treatments, but in root the difference was not found. On the whole, the three stresses lead to significant reduction in Ca accumulation in both roots and shoots relative to control, but

**Table 1 Interaction of NaCl and Cd on chlorophyll concentration, plant height and biomass in barley**

Genotypes	Treatments	SPAD value	Plant height (cm)	Root biomass (g/plant) (DW)	Shoot biomass (g/plant) (DW)
Gebeina	Control	42.03 a	58.9 a	0.21 a	0.47 a
	Cd	35.30 b	50.5 b	0.16 b	0.39 b
	NaCl	36.57 b	46.2 b	0.14 b	0.30 c
	NaCl+Cd	34.43 b	41.9 c	0.14 b	0.27 c
Zhou 1	Control	35.33 a	47.2 a	0.19 a	0.36 a
	Cd	30.27 b	41.8 ab	0.16 b	0.35 a
	NaCl	29.17 b	39.3 bc	0.12 c	0.25 b
	NaCl+Cd	27.00 b	36.6 c	0.11 c	0.24 b
Newgoutei	Control	39.13 a	49.2 a	0.20 a	0.38 a
	Cd	31.90 b	35.7 b	0.16 b	0.25 b
	NaCl	30.57 b	30.9 bc	0.10 c	0.18 c
	NaCl+Cd	30.23 b	27.3 c	0.09 c	0.16 c
Quzhou	Control	35.41 b	48.6 a	0.16 a	0.38 a
	Cd	28.90 b	43.5 a	0.15 a	0.31 b
	NaCl	30.50 b	30.1 b	0.09 b	0.20 c
	NaCl+Cd	22.93 c	30.1 b	0.08 b	0.19 c

Within each genotype, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's test)

**Table 2 Interaction of salinity and cadmium on K, Ca and Mg concentrations in barley**

Genotypes	Treatments	Concentration (mg/g DW)					
		Root			Shoot		
		K	Ca	Mg	K	Ca	Mg
Gebeina	Control	9.77 c	2.78 a	5.54 a	35.87 a	3.25 a	6.85 a
	Cd	12.06 b	2.07 b	5.07 a	30.04 b	2.68 b	6.11 a
	NaCl	15.63 a	1.99 b	1.55 b	32.16 b	1.64 c	4.03 b
	NaCl+Cd	12.76 b	1.88 c	1.09 b	30.69 b	1.51 c	3.50 b
Zhou 1	Control	8.18 b	2.54 a	3.92 a	33.19 a	5.26 a	10.41 a
	Cd	8.91 b	1.99 c	3.95 a	26.96 b	4.84 a	8.81 b
	NaCl	16.77 a	2.22 bc	1.33 b	30.99 a	1.79 b	4.07 c
	NaCl+Cd	15.02 a	2.35 b	1.13 b	22.32 c	1.74 b	3.75 c
Newgoutei	Control	10.45 ab	2.73 a	5.05 a	36.79 a	3.87 a	7.76 a
	Cd	12.17 a	2.04 b	4.97 a	29.94 b	2.74 b	6.84 b
	NaCl	11.57 ab	1.96 b	1.31 b	28.54 b	1.49 c	3.92 c
	NaCl+Cd	9.77 b	2.09 b	0.83 b	30.83 b	1.32 c	4.47 c
Quzhou	Control	11.58 b	2.54 ab	3.99 a	30.89 a	3.79 a	7.68 a
	Cd	13.03 ab	2.79 a	4.28 a	26.40 b	3.24 b	6.98 a
	NaCl	14.70 a	2.02 c	0.89 b	27.32 b	1.54 c	4.64 b
	NaCl+Cd	14.54 a	2.25 bc	0.95 b	27.33 b	1.32 c	4.68 b

Within each genotype, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's test)

**Table 3 Interaction of salinity and cadmium on K, Ca and Mg accumulation in barley**

Genotypes	Treatments	Accumulation (mg/plant)					
		Root			Shoot		
		K	Ca	Mg	K	Ca	Mg
Gebeina	Control	2.03 ab	0.58 a	1.15 a	16.75 a	1.52 a	3.21 b
	Cd	1.89 ab	0.33 b	0.79 b	11.69 b	1.04 b	2.37 c
	NaCl	2.26 a	0.27 bc	0.21 c	9.54 bc	0.49 c	1.19 e
	NaCl+Cd	1.72 b	0.25 c	0.15 c	8.27 c	0.40 c	0.94 ef
Zhou 1	Control	1.59 b	0.48 a	0.75 a	12.02 a	1.91 a	3.79 a
	Cd	1.48 b	0.33 b	0.65 b	9.47 b	1.70 b	3.10 b
	NaCl	2.01 a	0.27 b	0.16 c	7.81 b	0.45 c	1.03 ef
	NaCl+Cd	1.72 ab	0.27 b	0.13 c	5.25 c	0.41 c	0.88 ef
Newgoutei	Control	2.06 a	0.53 a	0.99 a	14.00 a	1.33 a	2.95 b
	Cd	1.81 a	0.30 b	0.73 b	7.49 b	0.69 b	1.72 d
	NaCl	1.20 b	0.20 c	0.14 c	5.05 c	0.26 c	0.69 f
	NaCl+Cd	0.92 b	0.20 c	0.08 c	5.07 c	0.22 c	0.74 f
Quzhou	Control	1.91 a	0.42 a	0.71 a	11.52 a	1.23 a	2.88 b
	Cd	1.95 a	0.39 a	0.64 a	8.20 b	1.01 b	2.18 c
	NaCl	1.34 b	0.18 b	0.08 b	5.35 c	0.30 c	0.91 ef
	NaCl+Cd	1.22 b	0.22 b	0.08 b	5.13 c	0.25 c	0.87 ef

Within each genotype, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's test)

for Zhou 1 there was no significant difference between Cd stress and the control (Table 3). Moreover, NaCl+Cd stress had lower root Ca accumulation than Cd stress alone for all genotypes except Zhou 1. However, no significant difference was found between NaCl and NaCl+Cd treatments. For shoot Ca

accumulation, significant difference was found between Cd and NaCl+Cd, but there was no significance between NaCl and NaCl+Cd treatments. On the whole, salt tolerant genotypes accumulated more Ca in roots and shoots than sensitive genotypes when exposed to NaCl and NaCl+Cd stresses.

Both NaCl alone and NaCl+Cd stresses led to significant decrease in root Mg concentrations for all barley genotypes (Table 2), while Cd stress had little effect on root Mg concentration. Both NaCl and NaCl+Cd stresses caused significant decrease in shoot Mg concentrations for all genotypes, but Cd stress had no significant effect on it. There was no significant difference between salt tolerant and sensitive genotypes under NaCl or combined stress. The three stress treatments, especially ones containing NaCl and NaCl+Cd, reduced significantly both root and shoot Mg accumulation for all barley genotypes (Table 3). Moreover, more reduction occurred in NaCl+Cd stress than Cd stress alone. The two salt tolerant genotypes had more Mg accumulation in roots than the two sensitive ones under NaCl and NaCl+Cd stresses.

#### **Micronutrient concentration and accumulation**

Cd stress significantly increased root and shoot Cu concentrations for all genotypes. However, NaCl stress alone did not affect root and shoot Cu concentrations for all genotypes, but for Newgoutei, which had significantly lower Cu concentration in NaCl treatment than in the control. NaCl+Cd treatment significantly increased shoot Cu concentration, but had no significant effect on root Cu concentration (Table 4). Zhou 1 had the highest root Cu concentration and another salt tolerant genotype Gebeina ranked the second under NaCl and NaCl+Cd stresses. The three stress treatments had little effect on root Cu concentration. Cd stress decreased root Cu accumulation in all genotypes, but the difference with the control was not significant (Table 5). NaCl and NaCl+Cd stresses decreased root Cu accumulation in salt sensitive genotypes, and had no significant effect on salt tolerant genotypes. Although NaCl and NaCl+Cd treatments caused marked reduction in shoot Cu accumulation of all genotypes, the two salt tolerant genotypes were relatively less affected than the two sensitive ones.

Root Fe concentration in all genotypes was decreased under Cd stress and increased under NaCl stress, compared with the control (Table 4). No significant difference was found in root Fe concentration between the combined stress and the control for all genotypes except Gebeina, which had significantly higher root Cd concentration than the control. Cd

stress decreased shoot Fe concentration in Zhou 1 but had little effect on the other three genotypes. NaCl and NaCl+Cd stresses caused significant increase in shoot Fe concentration for Quzhou and had little effect on other genotypes relative to the control. The three stresses decreased Fe accumulation in both roots and shoots of all genotypes (Table 5). Under Cd stress alone, Gebeina, a salt tolerant genotype, had lower root Fe accumulation than the other three genotypes. Yet under NaCl and NaCl+Cd stresses, salt tolerant genotypes Gebeina and Zhou 1 had higher root Fe accumulation than the two sensitive genotypes Newgoutei and Quzhou.

Cd stress showed little effect on Mn concentration in barley roots, while NaCl and NaCl+Cd stress decreased it significantly (Table 4). Cd stress decreased shoot Mn concentration in the salt tolerant genotypes and had little effect on the two salt sensitive genotypes. NaCl stress alone had no obvious effect on shoot Mn concentration, while NaCl+Cd stress caused marked reduction in Zhou 1 and Quzhou. The three stresses decreased root Mn accumulation in all genotypes, with NaCl+Cd stress having the greatest effect (Table 5). The two salt tolerant genotypes had higher root Mn accumulation than the two sensitive ones under NaCl and NaCl+Cd stresses. Under Cd stress alone, genotypic difference in root and shoot Mn accumulation was also detected. The three stresses decreased shoot Mn accumulation in all genotypes, with NaCl+Cd stress having the greatest effect. Under Cd stress, the two salt tolerant genotypes had significantly higher Mn accumulation than one salt sensitive genotype Newgoutei, but showed no difference with that in Quzhou. On the whole, salt tolerant genotypes had higher shoot Mn accumulation than sensitive ones when exposed to NaCl and NaCl+Cd stresses.

Cd stress decreased and NaCl stress increased root Zn concentration, respectively (Table 4). The effect of combined NaCl+Cd stress on Zn concentration varied with genotypes: Gebeina and Newgoutei showed decrease and Zhou 1 increase, and Quzhou showed little change in root Zn concentration. Cd stress decreased shoot Zn concentration in Zhou 1 and Newgoutei, and increased it in Gebeina. While for Quzhou, little change was found relative to the control. NaCl and NaCl+Cd stresses increased shoot Zn concentration in all genotypes, with the salt tolerant

**Table 4 Interaction of salinity and cadmium on Cu, Fe, Mn and Zn concentration in barley**

Genotypes	Treatments	Concentration ( $\mu\text{g/g DW}$ )							
		Root				Shoot			
		Cu	Fe	Mn	Zn	Cu	Fe	Mn	Zn
Gebeina	Control	38.9 b	360.6 b	75.57 c	30.20 a	18.84 b	167.3 c	18.59 b	10.83 c
	Cd	49.4 a	419.5 a	84.56 a	21.64 b	26.11 a	183.8 ab	27.64 a	19.99 a
	NaCl	34.3 b	381.4 b	77.60 bc	19.91 bc	18.48 b	173.1 bc	16.98 b	17.14 b
	NaCl+Cd	37.6 b	355.4 b	82.40 ab	18.53 c	24.80 a	190.9 a	16.26 b	11.05 c
Zhou 1	Control	39.1 b	273.7 c	74.14 b	19.12 a	18.65 b	162.1 b	12.29 c	13.56 a
	Cd	47.4 a	316.9 a	83.43 a	12.42 c	25.69 a	161.9 b	22.55 a	14.88 a
	NaCl	38.5 b	342.8 b	74.14 b	16.56 ab	18.77 b	159.1 b	15.98 b	12.09 b
	NaCl+Cd	40.6 ab	332.7 b	77.22 b	15.44 bc	26.20 a	191.4 a	16.26 b	11.76 b
Newgoutei	Control	62.5 b	527.8 ab	69.60 ab	37.91 a	20.48 c	183.4 b	18.31 b	29.98 b
	Cd	74.0 a	556.3 a	73.19 a	39.33 a	25.14 a	174.5 bc	26.10 a	32.84 a
	NaCl	50.3 c	531.5 ab	66.60 bc	26.07 c	17.21 d	160.7 c	18.69 b	24.67 c
	NaCl+Cd	58.2 b	512.3 b	62.94 c	30.41 b	24.34 a	220.5 a	14.70 c	24.72 c
Quzhou	Control	51.8 b	419.6 a	50.69 b	21.20 b	20.04 b	183.9 bc	16.21 a	25.45 a
	Cd	70.3 a	448.2 a	74.57 a	30.09 a	27.60 a	199.3 b	18.49 a	26.57 a
	NaCl	46.5 b	354.7 b	51.87 b	16.07 c	17.71 b	164.0 c	16.14 a	18.45 b
	NaCl+Cd	47.4 b	382.0 bd	53.73 b	16.68 c	27.61 a	228.2 a	12.69 b	16.90 b

Within each genotype, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's test)

**Table 5 Interaction of salinity and cadmium on Cu, Fe, Mn and Zn accumulations in barley**

Genotypes	Treatments	Accumulation ( $\mu\text{g/plant}$ )							
		Root				Shoot			
		Cu	Fe	Mn	Zn	Cu	Fe	Mn	Zn
Gebeina	Control	8.01 b	75.5 a	15.79 a	6.27 a	8.82 a	78.6 a	8.69 b	5.06 de
	Cd	9.42 a	80.1 a	16.16 a	5.48 b	9.15 a	66.7 bc	10.10 a	7.23 b
	NaCl	6.69 c	74.7 a	15.18 ab	4.70 c	7.25 b	65.6 bc	5.86 e	6.54 bc
	NaCl+Cd	6.18 c	58.3 b	13.51 b	4.18 d	4.70 c	71.6 b	5.29 ef	4.11 f
Zhou 1	Control	6.15 b	42.6 c	11.58 b	3.91 a	8.17 a	63.2 cd	4.80 fgh	5.27 d
	Cd	7.85 a	51.8 a	13.78 a	3.41 b	6.49 b	56.9 de	7.90 c	5.17 d
	NaCl	5.70 b	50.5 be	11.18 b	3.05 c	4.82 c	39.8 h	4.01 i	3.03 g
	NaCl+Cd	6.08 b	49.6 b	11.53 b	2.97 c	5.37 bc	59.9 cde	5.10 fg	3.68 fg
Newgoutei	Control	8.89 a	74.3 a	10.00 a	2.86 a	9.49 a	54.3 ef	5.41 ef	8.86 a
	Cd	8.89 a	66.9 b	8.73 ab	2.80 a	7.04 b	44.1 gh	6.60 d	8.25 a
	NaCl	5.23 b	55.3 c	6.92 b	2.70 a	3.04 c	28.4 i	3.30 j	4.37 ef
	NaCl+Cd	5.33 b	46.8 d	5.76 b	2.40 a	6.39 b	43.5 gh	2.90 jk	4.88 de
Quzhou	Control	6.99 b	56.5 a	6.82 ab	2.30 a	6.07 b	49.4 fg	4.40 ghi	6.89 b
	Cd	8.02 a	51.2 b	8.48 a	2.01 a	9.01 a	46.8 g	4.34 hi	6.24 c
	NaCl	4.38 c	33.2 c	4.88 b	1.51 b	2.92 c	27.0 i	2.65 jk	3.03 g
	NaCl+Cd	3.99 c	32.0 c	4.51 b	1.40 b	5.15 b	42.6 gh	2.38 k	3.13 g

Within each genotype, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's test)

genotypes having more increase than the sensitive ones. Both Cd and NaCl stresses decreased root Zn accumulation in all four genotypes, except for Zhou 1 in NaCl stress (Table 5). NaCl+Cd stress decreased root Zn accumulation in all genotypes, with the effect being more severe than Cd stress alone. Under

NaCl stress, the two salt tolerant genotypes had higher root Zn accumulation than the two salt sensitive ones. Cd and NaCl+Cd stresses decreased shoot Zn accumulation in all genotypes but Gebeina. Under all stresses, the two salt tolerant genotypes had higher shoot concentrations than the sensitive ones.

### Cd and Na concentration and accumulation

Cd concentration in both roots and shoots increased markedly when plants were exposed to Cd-containing medium, compared to the control (Table 6). However, NaCl+Cd treatment had markedly lower Cd concentration than Cd alone treatment, in particular for shoot Cd concentration of Zhou 1. There was a significant difference among the four genotypes in both root and shoot Cd concentrations. Hence, in the two treatments with Cd addition, a salt tolerant genotype Gebeina and a salt sensitive genotype Quzhou had the highest and lowest root Cd concentration, respectively. In terms of shoot Cd concentration, another salt-tolerant genotype Zhou 1 and a sensitive one Newgoutei ranked the highest and lowest, respectively. Regarding Cd accumulation, significant difference was found between Cd alone and NaCl+Cd treatments. Thus NaCl addition in the Cd-containing medium led to significant reduction of Cd accumulation in both roots and shoots. Similar to Cd concentration, there was a significant difference in Cd accumulation among genotypes. Zhou 1 had the highest shoot Cd accumulation in both Cd-containing treatments and root Cd accumulation in Cd alone treatment. While in NaCl+Cd treatment, Gebeina ranked the highest.

Compared to NaCl alone treatment, NaCl+Cd treatment increased Na concentration for all genotypes, but for Zhou 1 (Table 6). Moreover, the two salt tolerant genotypes (Gebeina and Zhou 1), on the whole, were lower than the two sensitive genotypes (Newgoutei and Quzhou), in particular for shoots. In terms of Na accumulation, the effect of Cd stress varied with genotypes. Hence, Gebeina had more Na accumulation of both root and shoot in NaCl+Cd treatment than in NaCl alone treatment, while Newgoutei had less root Na accumulation in the NaCl+Cd stress than in NaCl stress alone.

### DISCUSSION

Excessive Cd and/or NaCl accumulation affects the rate of uptake and distribution of certain essential nutrients in plants, and consequently may be responsible for mineral deficiencies/imbalance and depression of the plant growth (Rubio *et al.*, 1994; Hernandez *et al.*, 1996; Zhang *et al.*, 2002; Dražić *et al.*, 2004). In the present study, NaCl and Cd stresses caused significant decline in chlorophyll concentration, plant height and biomass in all genotypes. Nevertheless, the combined stress (NaCl+Cd) did not lead

**Table 6 Interaction of salinity and cadmium on Cd and Na concentration and accumulations in barley**

Genotypes	Treatments	Concentration ( $\mu\text{g/g DW}$ )				Accumulation ( $\mu\text{g/plant}$ )			
		Root		Shoot		Root		Shoot	
		Cd	Na	Cd	Na	Cd	Na	Cd	Na
Gebeina	Control	0.007 b	1.41 c	0.005 b	1.59 c	0.0015 b	0.30 c	0.0018 b	0.74 c
	Cd	35.650 a	1.52 c	3.230 a	1.09 c	4.1700 a	0.24 c	1.2500 a	0.43 c
	NaCl	0.013 b	43.95 b	0.007 b	76.57 b	0.0018 b	6.27 b	0.0027 b	22.54 b
	NaCl+Cd	30.770 a	65.79 a	3.690 a	101.68 a	3.6200 a	8.88 a	0.9900 a	27.35 a
Zhou 1	Control	0.013 b	0.88 b	0.002 c	0.97 b	0.0025 c	0.17 c	0.0017 c	0.36 b
	Cd	28.800 a	0.98 b	11.240 a	0.78 b	4.6300 a	0.16 c	3.9300 a	0.28 b
	NaCl	0.014 b	46.15 a	0.007 c	104.69 a	0.0017 c	5.46 b	0.0026 c	26.46 a
	NaCl+Cd	26.870 a	54.54 a	8.240 b	113.43 a	2.7300 b	6.19 a	1.9300 b	26.63 a
Newgoutei	Control	0.011 b	0.94 b	0.002 b	0.99 b	0.0021 c	0.18 b	0.0015 b	0.38 b
	Cd	32.520 a	1.07 b	3.080 a	0.92 b	4.4700 a	0.16 b	0.7800 a	0.23 b
	NaCl	0.014 b	53.20 a	0.007 b	113.93 ad	0.0015 c	5.51 a	0.0012 b	20.14 a
	NaCl+Cd	29.810 a	44.96 a	2.800 a	125.87 a	2.7900 b	4.20 a	0.4600 a	20.74 a
Quzhou	Control	0.005 b	1.48 c	0.003 b	1.96 b	0.0008 c	0.24 c	0.0012 b	0.73 b
	Cd	24.970 a	1.87 c	3.510 a	1.67 b	3.7300 a	0.28 c	1.0900 a	0.52 b
	NaCl	0.013 b	49.95 b	0.006 b	134.41 a	0.0012 c	4.57 b	0.0010 b	26.50 a
	NaCl+Cd	23.380 a	88.26 a	4.170 a	139.39 a	1.9600 b	7.43 a	0.7800 a	25.99 a

Within each genotype, different letters indicate significant differences at  $P \leq 0.05$  (Duncan's test)

to additional effect, that is, the decreased value of NaCl+Cd stress did not approximate to the total decreased value of NaCl and Cd stresses.

The studies reported to date about the effect of Cd toxicity on nutrient uptake and accumulation in plants sorption have provided contradicting results, which were presumably due to the differences in the culture methods, species, organs, and conditions such as concentration in medium, growth period and temperature. Liu *et al.* (2003b) reported that significant positive correlations between Cd and Fe, Cd and Zn, Cd and Cu existed in rice in terms of their concentrations in roots and leaves. In contrast, Metwally *et al.* (2005) indicated that toxic Cd levels inhibited uptake of nutrient elements such as P, K, S, Ca, Zn, Mn, and B by plants in an organ- and genotype-specific manner in pea. Cd toxicity also affected concentration of some nutrients in barley (Wu and Zhang, 2002; Wu *et al.*, 2003). In the current study, we found that Cd stress reduced Ca and Mg concentrations in roots and shoots, K concentrations in shoots, increased K and Cu concentrations in roots relative to control, but had non-significant effect on Cu, Fe and Mn concentrations in shoot.

On the other hand, salinity stress may also lead to nutritional disorders in crops because of the effect of salinity on nutrient availability, competitive uptake, transport or partitioning within the plant. However, the reports available up to date were mainly focused on the effect of salinity on  $\text{Ca}^{2+}$  and  $\text{K}^+$  (Lynch and Lauchli, 1985; Gorham, 1990; Wolf *et al.*, 1991; Wei *et al.*, 2003). In our research, Cd, NaCl and NaCl+Cd stresses decreased Ca and Mg concentration, and increased K and Cu concentration in roots of all barley genotypes as compared to the control. The three stresses resulted in marked reduction in shoot Ca, K and Mg concentration, and had no significant effect on Cu, Fe and Mn concentration. Moreover, the effect of Cd, NaCl and NaCl+Cd stresses on shoot Zn concentration varied with the stress type and genotype, and the present study provided some results different from those reported by Jalil *et al.* (1994) who found that Cd application decreased the concentration of K, Zn, and Mn in roots and shoots of durum wheat, while the Fe and Cu concentrations in shoots and roots were not affected. The three stresses reduced most nutrient accumulation in both roots and shoots, which was similar to the results of Yang *et al.* (1998) who re-

ported that addition of Cd to growth medium decreased the accumulation of Fe, Mn, Cu, Ca, and Mg in cabbage, ryegrass, maize and white clover. Salinity and NaCl+Cd stresses increased root K and shoot Cu concentration in the two salt tolerant genotypes. In conclusion, the pattern of Cd stress effect on nutrient uptake and accumulation was different from that of salinity stress, and the effect of NaCl+Cd stress does not mean the simple pool of Cd and NaCl stress alone, i.e. complex interaction occurs when the two stresses were exposed simultaneously.

Salinity has been identified as an important determinant of Cd concentration in crops (Smolders and McLaughlin, 1996; Helal *et al.*, 1999; Weggler-Beaton *et al.*, 2000) although the mechanism has not yet been elucidated. It was reported that Cd uptake was enhanced when plants grew in soils with higher NaCl content (McLaughlin *et al.*, 1994; Li *et al.*, 1994). Although the detailed interaction between salinity and heavy metals, including Cd is still not fully understood, Weggler-Beaton *et al.* (2000) assumed that the effect of salinity in increasing Cd bio-availability is attributable to the formation of Cd-Cl complexes, predominantly the 1:1 ( $\text{CdCl}^+$ ) and 1:2 ( $\text{CdCl}_2$ ) complexes. These complexes are less strongly sorbed to soil than free  $\text{Cd}^{2+}$  ion and hence increase Cd mobility at the soil-root interface. Moreover, these complexes also stimulate transport of Cd across the zone encompassing soil-rhizosphere-apoplast-plasma membrane. Thus, increased soil-plant transfer of Cd can occur under salinity. In the present research, nevertheless, we found that addition of NaCl in Cd stressed solution resulted in decrease of Cd concentration in both roots and shoots. The reason for the inconsistency between our results and those of others could be attributable to the different culture conditions. In the experiments with soil media, enhanced Cd uptake was due to the fact that  $\text{Cl}^-$  forms complexes with Cd sorbed to soil originally. While in our experiment using hydroponic solution, Cd is fully dissolved, so formation of Cl-Cd complexes has no distinct effect on Cd bio-availability. Meanwhile, it might be possible that injured roots due to high salinity weaken the capacity of ion uptake, or Na ion competitively inhibits Cd uptake. The result being different from the hydroponic research of Mühling and Läuchli (2003) was probably due to the NaCl concentration and pH.



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