Front. For. China 2009, 4(2): 208–215
DOI 10.1007/s11461-009-0007-5

RESEARCH ARTICLE

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Effect of NaCl stress on ion distribution in roots and growth of *Cyclocarya paliurus* seedlings

Abstract We studied ion distribution in roots and the growth of *Cyclocarya paliurus* seedlings of three provenances, Huangshan in Anhui, Jiujiang in Jiangxi and Kunming in Yunnan, under conditions of 0, 1, 3 and 5 g/L NaCl stress using X-ray microanalysis. Results show that under NaCl stress of 3 and 5 g/L, the relative contents of Na^+ and Cl^- in root tissues increased, while the relative contents of K^+, Ca^{2+} and Mg^{2+} decreased. With an increase in salinity, the relative content of Na^+ in the epidermis and cortex of the root increased, while the relative content of Cl^- in the stele and cortex of the root increased markedly. Thus, ions in the root tissues were unbalanced and the ratios K^+/Na^+ and Ca^{2+}/Na^+ decreased, while $Na^+/(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$ increased. The decrease of the K^+/Na^+ ratio and the substantial increase of Cl^- in root tissues contributed to a decline in seedlings survival and reduced the increments for seedling leaf area, height, basal diameter as well biomass. Our preliminary conclusion is that the level of salt tolerance for the tested provenance seedlings was in the order of Huangshan > Kunming > Jiujiang, and the threshold of salt tolerance for *C. paliurus* seedlings was about 1 g/L.

Keywords NaCl stress, *Cyclocarya paliurus*, ion distribution, root tissue, X-ray microanalysis

1 Introduction

There are about 380 million hm^2 of potentially usable areas for agriculture in the entire world. However, in many of these areas, production has been severely restricted by salinity (Lambers, 2003). Besides a decreased water potential, excessive Na^+ and Cl^- was also discovered as a key factor limiting plant growth in the case of salt stress. These harmful mineral elements are not required by most glycophytes for normal growth (Niu et al., 1995). High amounts of Na^+ and Cl^- in tissues are, therefore, often considered as the most critical factors responsible for salt toxicity in non-halophytes (Greenway and Munns, 1980; Niu et al., 1995). Ward et al. (2003) indicated that growing plants with deep roots would provide a long-term solution for managing the dryland salinity problem. Considerable efforts are being made to develop salt-tolerant cultivars of plant species which, when integrated into appropriate management programs, may allow the exploitation of saline soils. If parallels were drawn with early selection and breeding programs to improve crop species for non-saline environments, we would clearly observe that very significant and rapid advancement in performance in a wide range of crop species could be achieved through the exploitation of considerable useful genetic variability present within those crops in their relatively unimproved state (Ashraf and Mcneilly, 1987; Morabito et al., 1996).

Cyclocarya paliurus (Batal) Iljinskaja (family Juglandaceae), native to China, is the sole species in its genus and mainly grows at 420–2500 m elevation in mountainous regions. In China, the bark and leaves of *C. paliurus* are widely used to produce medicinal tea (Xie and Li, 2001). A new crystalline compound, called cyclocaric acid A (3, 23-β-epoxy-olean-12-en-28-oicacid), was isolated from the leaves of *C. paliurus* (Fang and Fu, 2007) and a rich polysaccharide content from leaves of *C. paliurus* was confirmed, which is effective in reducing blood glucose and improving the capacity of glucose tolerance in diabetic mice (Xie and Li, 2001; Li et al., 2002). An *in vitro* study shows that *C. paliurus* inhibits alpha-glucosidase and a disaccharide-degrading enzyme in the small intestinal mucosa, leading to a decrease in the absorption of glucose into the blood and a subsequent lowering of the blood glucose level (Kurihara et al., 2003). Xie and Li (2001) reported that the contents of flavonoid, vitamin E (VE) and vitamin C (VC) in the leaves of *C. paliurus* were also higher than in other plants. A huge production of tender leaves is required as raw material from *C. paliurus* tea and

Translated from *Scientia Silvae Sinica*, 2008, 44(6): 66－72 [译自: 林业科学]

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medical use. Greater demands for leaf production will be placed on new plantations of *C. paliurus*. There are, in total, about 27×10^6 hm^2 saline soil areas in China, of which coastal lands account for 8% (Fang et al., 2006). Therefore these land resources would be a great potential for developing forestry. At present, there is no information available on the salt tolerance of *C. paliurus* at different stages of its life cycle. Roots are the important interface between plants and the environment. When environments alter, the roots of plants will be affected first. Then, roots perceive adverse signals and contribute to corresponding physiological changes. Peng et al. (2004) reported that plant growth is constrained due to the loss of balance of ion distributions in roots under salt stress. Since the mechanism by which plants tolerate salt is complex and differs from species to species (Greenway and Munns, 1980; Ashraf and Harris, 2004), the objectives of our study are: (a) to investigate the ion distribution and radial transport in root tissues using X-ray microanalysis and (b) to evaluate the salt-tolerance of *C. paliurus* seedlings. The results from this study will contribute to an enhanced understanding of the salt-tolerance mechanism of *C. paliurus* seedlings and provide some scientific and fundamental bases for site selection and plantation silviculture.

2 Materials and methods

2.1 Plant material and growth conditions

Seeds of *C. paliurus* were collected in October 2005 from Huangshan, Anhui Province (H), Jiujiang, Jiangxi Province (J) and Kunming, Yunnan Province (K), China. Seed trees were about 15 m tall and 28 cm in diameter at breast height. After drying in open sunlight and removal of unwanted material, the seeds were treated with concentrated sulphuric acid and flushed seeds over. The seeds were stratified in pails according to a size ratio of 1 (seed) to 3 (sand, mixed with giberrellin) under natural conditions in order to break seed dormancy. The stratification was started in January 2006 until March 2006. Seeds were sown in containers with pearl stones, vermiculite and peat soil (1∶2∶2) after they were stratified. When the height of the seedlings was about 7 cm, they were transplanted into black plastic boxes with a 1/2 strength Hoagland nutrient solution for cultivation of seedlings growth adaptability (Pan, 1995). The dimensions of the 8 mm thick plastic boxes were 56 cm (length)×36 cm (width)×26 cm (height). After 10 d of cultivation, uniform seedlings were selected and placed in plastic boxes filled with the Hoagland nutrient solution containing the different NaCl treatments. The salt treatment was started in July 2006 with four levels of NaCl, i.e., 0 (CK), 17.0 (S1), 51.0 (S2) and 85.0 (S3) mol/L of NaCl (equivalent to 0, 1.0, 3.0 and 5.0 g/L of NaCl). There were about 45 seedlings for each treatment of any provenance. The solutions were aerated throughout the experiment. The volumes were maintained by adding water to compensate for water loss by evaporation and transpiration and the nutrient solutions renewed every seven days since the seedlings were transplanted to the boxes. The seedlings were grown at a photon flux density of 350–400 μmol/(m^2·s) with 12 h sunshine, $26\pm0.5/20\pm0.5$°C temperature and 70/80% relative humidity in a day/night cycle in the phytotron (type RGS-20).

2.2 X-ray microanalysis

After 23 d of salt treatment, the roots of seedlings from the different treatments were washed with distilled water three times. The harvested root segments, including the tip and 3 cm or more of the root, were immediately sliced free-hand with a razor blade to obtain transverse sections. The samples were dried naturally, carbon-coated in a high vacuum sputter coater and stored in a desiccator (van Steveninck and van Steveninck, 1991). Samples were analyzed in a JSM-6300 scanning electron microscope equipped with an energy-dispersive X-ray detector (Sigma) (Tomos et al., 1994). Counts per second of [Na^+], [K^+], [Ca^{2+}], [Mg^{2+}] and [Cl^-] were measured in roots from different treatments. Four tissue samples, i.e. epidermal cells, cortical cells and the stelar parenchyma in each root transverse section were analyzed. More than three transverse sections of each treatment were observed and three locations of the same tissue of each section were analyzed. Both map- and line-scans were carried out (Li et al., 1991, 1996; Lindsay et al., 1996).

2.3 Measurements of seedlings growth

The effects of the four concentrations of NaCl on seedling growth indices of *C. paliurus* were examined in the phytotron. Survival, leaf area, height, basal diameter and biomass of the seedlings were measured once before salt treatment and 28 d after the treatment. More than three plants of each treatment were taken for biomass measurement. Then their dry weights (dried at 70°C for 24 h) were measured (Fang et al., 2006).

2.4 Data analysis

Data from our study were analyzed using SPSS 11.5 (Statistical Product and Service Solutions) software. After conducting an analysis of variance (ANOVA), the Duncan test was used to detect significant differences among the treatments with a probability of 95% ($\alpha = 0.05$).

3 Results

3.1 Ion contents and distribution in root tissues

Salt stress significantly affected root ion micro-distribution of *C. paliurus* seedlings (Fig. 1). Salt treatments induced

increases of Na^+ and Cl^- in root micro-tissues and Cl^- increase was more evident with the increase of NaCl concentration, while K^+, Ca^{2+} and Mg^{2+} levels were reduced. Under the 17.0 mol/L NaCl treatment, the trend in the variation of the relative ion content in the root microscope approached that of the control. This shows that the threshold of tested seedlings enduring salt stress was about 1.0 g/L. For 85.0 mol/L NaCl-treated seedlings of different provenances, the Na^+ and Cl^- relative contents of root microscope in the Jiujiang provenance from Jiangxi seedlings were 2.21 and 1.75 times larger than that of the Hunangshan provenance from Anhui seedlings and 1.40 and 1.22 times that of the Kunming provenance from Yunnan seedlings (Fig. 1), while the K^+ relative content of root microscope in the Jiujiang provenance from Jiangxi seedlings was 0.09 and 0.13 times that of the Huangshan provenance from Anhui and Kunming provenance from Yunnan seedlings. This shows that the capacity of salt tolerance for the Huangshan provenance was stronger than that of the Jiujiang provenance under heavy salt stress.

3.2 Ratios of ions [K^+/Na^+, Ca^{2+}/Na^+, $Na^+/(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$] in root tissues

Under salt treatment, with the increase of salinity, both ratios of K^+ to Na^+ and Ca^{2+} to Na^+ increased, while ratios of Na^+ to $(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$ decreased in the root microscope (Table 1). The result show that Na^+ evidently inhibited roots from absorbing K^+, Ca^{2+} and Mg^{2+} for 85 m NaCl-treated seedlings, so that the levels of nutrition were unbalanced. Generally, the ratios of K^+ to Na^+ and Ca^{2+} to Na^+ in the root microscope of the Jiujiang provenance from Jiangxi seedlings were lower, while ratios of Na^+ to $(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$ were higher than the in other two provenances. Meanwhile, the mean ratios of K^+ to Na^+, Ca^{2+} to Na^+ and Na^+ to $(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$ in the root microscope of the Jiujiang provenance from Jiangxi seedlings were 2.08, 1.15 and 2.14, while the Huangshan provenance from Anhui seedlings were 6.28, 8.64 and 0.69. This indicates that the degree of nutritional imbalance for the Jiujiang provenance from Jiangxi seedlings was more serious, i.e., the degree of salt toxicity was more evident.

3.3 Selective transportation of ions in root tissues

With the increase in salt concentration, Cl^- from the cortex into the stele of seedling roots increased. The Na^+ ratio of cortex to epidermis varied slightly, while the ratio of stele to cortex decreased. The variation for K^+ ratio of cortex to epidermis was slight, while the ratio of stele to cortex gradually increased. The Ca^{2+} ratio of cortex to epidermis decreased, while the changes in the Ca^{2+} ratio of stele to cortex were not apparent. The Mg^{2+} ratio of cortex to epidermis increased, while the Mg^{2+} ratio of stele to cortex decreased (Table 2). These results show that the damage

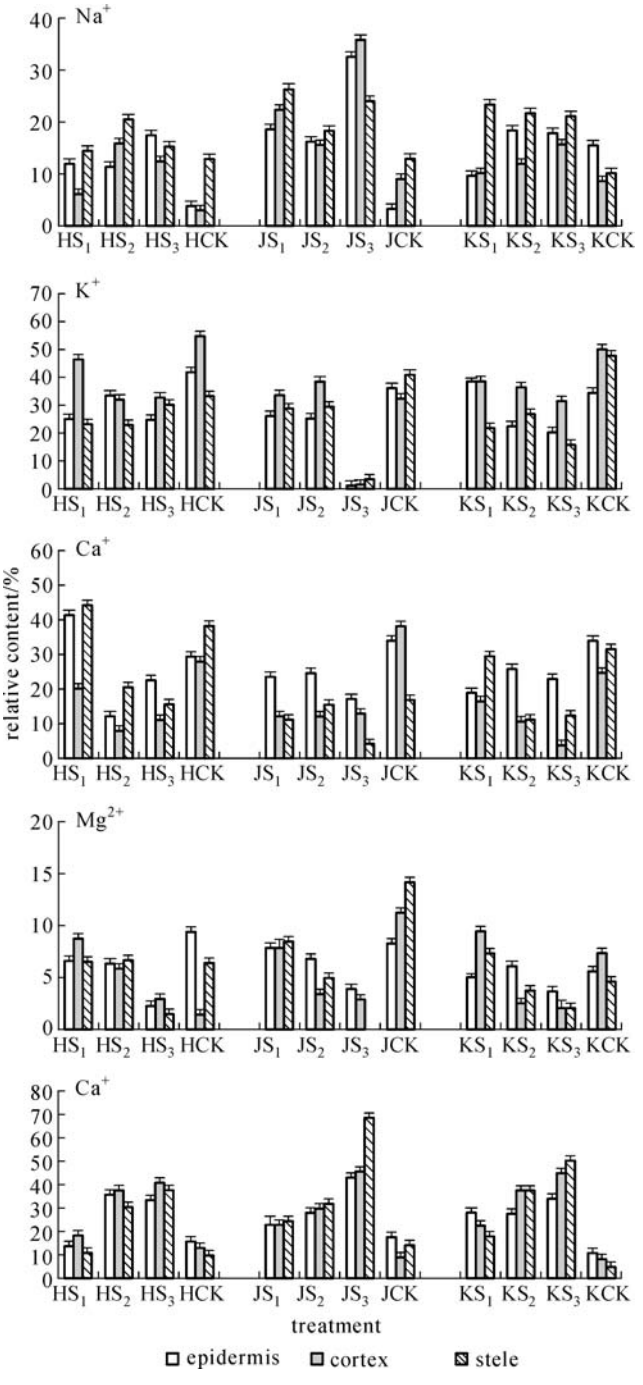

Fig. 1 Ion distribution in the root tissues after 23 days of salt stress.
Note: H, J, K show Huangshan, Jiujiang, Kunming provenances, while CK, S_1, S_2, S_3 show 0, 17.0, 51.0 and 85.0 mol/L NaCl treatments, respectively.

resulting from Cl^- toxicity was quicker and more serious than Na^+ toxicity under salt stress. Therefore, the higher the salinity, the greater the Cl^- ratio of stele to cortex and the more serious the degree of toxicity from which the seedling suffered under salt stress. For the three tested provenances, the difference among provenances was significant. Na^+ and

Table 1 Ion ratios in root tissues after 23 days of salt stress

tissue	treatment	Huangshan			Jiujiang			Kunming		
		K^+/Na^+	Ca^{2+}/Na^+	$Na^+/(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$	K^+/Na^+	Ca^{2+}/Na^+	$Na^+/(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$	K^+/Na^+	Ca^{2+}/Na^+	$Na^+/(K^+ + Na^+ + Ca^{2+} + Mg^{2+})$
epidermis	S_1	2.11 ± 0.83aBC	3.51 ± 6.49aA	0.14 ± 0.06bA	1.39 ± 0.39bB	1.27 ± 0.08cB	0.25 ± 0.11aB	3.94 ± 0.61aA	1.94 ± 0.33bAB	0.13 ± 0.04bB
	S_2	3.04 ± 0.96aB	1.13 ± 0.43B	0.17 ± 0.04bA	1.47 ± 0.80bB	1.53 ± 0.47B	0.23 ± 0.06aB	1.19 ± 0.06bB	1.46 ± 0.10AB	0.25 ± 0.11aA
	S_3	1.42 ± 0.16aC	1.27 ± 0.29aB	0.26 ± 0.02bA	0.06 ± 0.01bC	0.55 ± 0.25bC	0.58 ± 0.13aA	1.14 ± 0.42aB	1.31 ± 0.08aB	0.27 ± 0.16bA
	CK	11.11 ± 3.17aA	7.62 ± 2.38aA	0.05 ± 0.01bB	12.3 ± 1.98aA	12.04 ± 2.24aA	0.04 ± 0.01bC	2.28 ± 0.35bA	2.24 ± 0.26bA	0.17 ± 0.07aB
cortex	S_1	7.55 ± 3.56aB	3.32 ± 0.13aA	0.08 ± 0.03cB	1.53 ± 0.09bB	0.55 ± 0.05bB	0.29 ± 0.12aB	3.96 ± 0.80aAB	1.74 ± 1.29aA	0.13 ± 0.03abB
	S_2	2.02 ± 0.31C	0.51 ± 0.10B	0.26 ± 0.12A	2.59 ± 1.76AB	0.80 ± 0.19B	0.22 ± 0.05B	3.04 ± 0.66AB	0.93 ± 0.32B	0.19 ± 0.09B
	S_3	2.62 ± 1.93aBC	0.87 ± 0.25B	0.21 ± 0.05bA	0.06 ± 0.01bC	0.38 ± 0.06bB	0.66 ± 0.17aA	2.00 ± 0.94aB	0.31 ± 0.01B	0.29 ± 0.15bA
	CK	20.60 ± 4.40aA	10.49 ± 0.62aA	0.03 ± 0.01bC	3.75 ± 0.60cA	4.41 ± 1.48bA	0.10 ± 0.04aC	6.01 ± 3.99bA	3.07 ± 0.15cA	0.09 ± 0.02aC
stele	S_1	1.65 ± 1.06A	3.10 ± 1.90aA	0.16 ± 0.10bB	1.08 ± 0.64B	0.44 ± 0.05cB	0.35 ± 0.07aB	0.95 ± 0.27BC	1.29 ± 0.67bB	0.28 ± 0.12abBC
	S_2	1.10 ± 0.24B	1.00 ± 0.19B	0.29 ± 0.02A	1.62 ± 0.82AB	0.87 ± 0.27B	0.27 ± 0.03B	1.23 ± 0.14B	0.51 ± 0.12C	0.35 ± 0.14AB
	S_3	2.01 ± 0.07aA	1.00 ± 0.32aB	0.24 ± 0.04bA	0.15 ± 0.01cC	0.19 ± 0.03bC	0.75 ± 0.35aA	0.78 ± 0.47bC	0.59 ± 0.09aC	0.41 ± 0.21bA
	CK	2.61 ± 1.39A	3.04 ± 2.84aA	0.14 ± 0.01B	3.19 ± 0.81A	1.37 ± 0.08bA	0.15 ± 0.08C	4.82 ± 5.18A	3.21 ± 0.79aA	0.11 ± 0.04C

Note: Data were analyzed by Duncan's multiple range test; means followed by different letters were statistically different ($\alpha = 0.05$), where different small letters show significant difference among provenances for each NaCl treatment and different capital letters show significant difference among treatments for each provenance. The same comments apply to Table 2.

Table 2 Ion ratios of cortex to epidermis and stele to cortex in roots after 23 days of salt stress

ion	treatment	Huangshan		Jiujiang		Kunming	
		cortex/epidermis	stele/cortex	cortex/epidermis	stele/cortex	cortex/epidermis	stele/cortex
Na^+	S_1	0.52 ± 0.14bB	2.33 ± 0.63AB	1.20 ± 0.32aB	1.17 ± 0.24A	1.02 ± 0.16aA	2.30 ± 0.35
	S_2	1.44 ± 0.21aA	1.29 ± 0.54B	0.93 ± 0.21bB	1.19 ± 0.29A	0.65 ± 0.19bBC	1.81 ± 0.37
	S_3	0.72 ± 0.32bB	1.20 ± 0.54aB	1.10 ± 0.35aB	0.67 ± 0.15bB	0.90 ± 0.14abAB	1.30 ± 0.62a
	CK	0.70 ± 0.21bB	4.79 ± 1.26aA	3.01 ± 1.02aA	1.46 ± 0.65bA	0.55 ± 0.21bC	1.20 ± 0.15b
K^+	S_1	1.85 ± 0.28	0.51 ± 0.04bB	1.31 ± 0.52	0.83 ± 0.15aB	1.03 ± 0.22	0.55 ± 0.04bB
	S_2	0.95 ± 0.28b	0.70 ± 0.05AB	1.63 ± 0.65a	0.75 ± 0.21B	1.65 ± 0.29a	0.73 ± 0.17AB
	S_3	1.32 ± 0.65	0.92 ± 0.24bA	1.13 ± 0.35	1.53 ± 0.27aA	1.57 ± 0.25	0.50 ± 0.12bB
	CK	1.31 ± 0.52a	0.61 ± 0.32bB	0.92 ± 0.24b	1.24 ± 0.66aA	1.44 ± 0.19a	0.96 ± 0.31abA
Ca^{2+}	S_1	0.49 ± 0.21bB	2.17 ± 0.65a	0.52 ± 0.24bB	0.93 ± 0.16bA	0.92 ± 0.06aA	1.70 ± 0.37abAB
	S_2	0.64 ± 0.34B	2.53 ± 1.20a	0.48 ± 0.16B	1.30 ± 0.27abA	0.41 ± 0.14B	0.98 ± 0.06bB
	S_3	0.50 ± 0.08aB	1.37 ± 0.35a	0.76 ± 0.16aB	0.34 ± 0.11bB	0.21 ± 0.04bB	2.47 ± 0.24aA
	CK	0.96 ± 0.21abA	1.39 ± 0.52a	1.10 ± 0.29aA	0.45 ± 0.14bB	0.75 ± 0.19bA	1.25 ± 0.21aAB
Mg^{2+}	S_1	1.34 ± 0.57A	0.73 ± 0.21BC	1.00 ± 0.05AB	1.09 ± 0.33A	1.85 ± 0.69A	0.80 ± 0.11AB
	S_2	0.93 ± 0.24aA	1.12 ± 0.32B	0.49 ± 0.16bC	1.47 ± 0.56A	0.42 ± 0.14bB	1.18 ± 0.37A
	S_3	1.20 ± 0.56aA	0.52 ± 0.14aC	0.73 ± 0.27bBC	0.00 ± 0.01bB	0.59 ± 0.15bB	0.92 ± 0.35aAB
	CK	0.16 ± 0.05bB	4.26 ± 1.65aA	1.37 ± 0.34aA	1.26 ± 0.49bA	1.31 ± 0.24aA	0.60 ± 0.16cB
Cl^-	S_1	1.25 ± 0.37a	0.62 ± 0.21	0.98 ± 0.21abA	1.09 ± 0.44	0.83 ± 0.32b	0.77 ± 0.27
	S_2	1.05 ± 0.21	0.79 ± 0.05	1.09 ± 0.34A	1.06 ± 0.64	1.44 ± 0.62	1.00 ± 0.47
	S_3	1.22 ± 0.38	0.93 ± 0.54	1.05 ± 0.04A	1.50 ± 0.03	1.29 ± 0.29	1.09 ± 0.28
	CK	0.82 ± 0.24a	0.66 ± 0.27b	0.44 ± 0.16bB	1.84 ± 0.87a	0.81 ± 0.22a	0.57 ± 0.07b

Cl^- root micro-transportation were more active for NaCl-treated seedlings of the Jiujiang provenance from Jiangxi. Thus, the relative contents of Na^+ and Cl^- in the root microscope were higher and its mean ion ratio of stele to cortex was the least at only 1.06.

3.4 Effect of NaCl on seedling growth

The seedlings of *C. paliurus* had optimal survival and growth in the nutrient solution without any added NaCl (Fig. 2, Table 3). Low levels of salinity (17.0 mol/L of

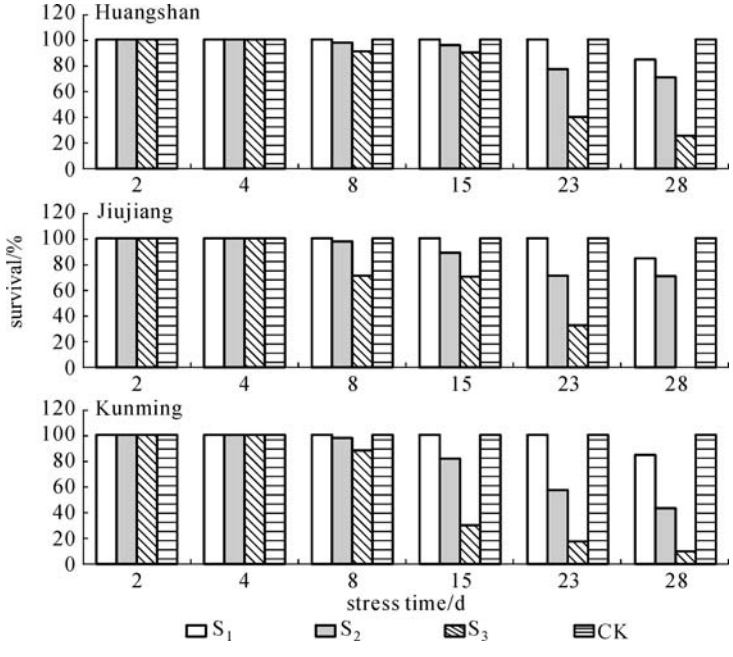

Fig. 2 Survival percentage of seedlings under salt stress

Table 3 Analysis of variance of seedling growth after 28 days of salt stress

index	treatment	Huangshan	Jiujiang	Kunming
leaf area increment/cm^2	S_1	5.34 ± 1.23aB	2.41 ± 0.36bB	2.25 ± 0.29bB
	S_2	0.07 ± 0.05C	–1.72 ± 0.46C	–0.22 ± 0.07C
	S_3	–1.22 ± 0.57C	–2.06 ± 0.58C	–2.45 ± 0.09C
	CK	11.63 ± 2.35aA	11.85 ± 3.29aA	8.57 ± 1.28bA
height increment/cm	S_1	3.10 ± 0.89AB	1.90 ± 0.55B	1.55 ± 0.29B
	S_2	1.20 ± 0.09aB	0.10 ± 0.02bC	0.35 ± 0.14bC
	S_3	0.10 ± 0.06C	0.05 ± 0.03C	0.15 ± 0.03C
	CK	5.10 ± 1.59bA	5.50 ± 1.69bA	8.75 ± 2.49aA
basal diameter increment/mm	S_1	0.83 ± 0.20aA	0.52 ± 0.24abB	0.41 ± 0.30bAB
	S_2	0.14 ± 0.11B	0.21 ± 0.13C	0.12 ± 0.05BC
	S_3	0.03 ± 0.02B	0.02 ± 0.01C	0.02 ± 0.01C
	CK	1.10 ± 0.33aA	1.24 ± 0.22aA	0.63 ± 0.22bA
biomass increment per plant/g	S_1	1.05 ± 0.12aA	0.11 ± 0.05bB	0.42 ± 0.26bAB
	S_2	0.44 ± 0.22aB	0.03 ± 0.01bC	0.08 ± 0.06bC
	S_3	0.22 ± 0.04aB	–0.21 ± 0.11bD	0.17 ± 0.12aBC
	CK	1.51 ± 0.06aA	1.22 ± 0.49abA	0.71 ± 0.35bA

NaCl) did not cause substantial inhibition of growth but the general tendency was that increasing concentrations of NaCl induced a progressive decline both in survival and seedling growth. There was a significant reduction in survival and increments in leaf area, height, basal diameter and biomass with increasing NaCl concentration (Fig. 2, Table 3).

Twenty eight days after the salt treatment, the survival of tested provenance seedlings in CK was 100%, while under 17.0, 51.0 and 85 mol/L NaCl treatment, seedling survival of the Huangshan provenance from Anhui was 85%, 70% and 25%, seedling survival of the Jiujiang provenance from Jinagxi was 84%, 33% and 0% and seedling survival of the Kunming provenance from Yunnan was 90%, 50% and 8%, respectively.

For the surviving seedlings of the three tested provenances, after 28 d of salt stress, the leaf area increments of the S_1, S_2 and S_3 treatments were 45.9%, 0.6% and –10.5% (Huangshan provenance from Anhui), 20.3%, –14.5% and –17.4% (Jiujiang provenance from Jiangxi), 26.3%, –2.6% and –28.6% (Kunming provenance from Yunnan). For the CK, seedling height increments were 60.8%, 23.5% and 1.96% (Huangshan provenance from Anhui), 34.5%, 1.8% and 0% (Jiujiang provenance from Jiangxi), 17.7%, 4.0% and 1.7% (Kunming provenance from Yunnan). For the CK, the basal diameter increments were 72.7%, 9.1% and 0% (Huangshan provenance from Anhui), 41.7%, 16.7% and 0% (Jiujiang provenance from Jiangxi), 66.7%, 16.7% and 0% (Kunming provenance from Yunnan) and the biomass increments for the CK were 69.5%, 29.1% and 14.6% (Huangshan provenance from Anhui), 15.5%, 4.2% and –29.6% (Jiujiang provenance from Jiangxi), 290.5%, 19.0% and 40.5% (Kunming provenance from Yunnan) of the CK, respectively.

4 Discussion

The results of our experiment indicates that root ion microvariation and growth of *C. paliurus* seedlings were clearly affected by the presence of NaCl, and significantly inhibited when NaCl concentration reached 85.0 mol/L. We found that after 23 d of 17.0 mol/L NaCl treatment, the salt treatment affected the tested *C. paliurus* seedlings growth. Most of the tested *C. paliurus* seedlings still could grow and develop and, with the exception of the Jiujiang provenance from Jiangxi, the growth conditions of the other two provenances seedlings approached that of the control. Relatively, the ability of tested provenance seedlings enduring salt stress was in the order of the Huangshan provenance from Anhui > the Kunming provenance from Yunnan > the Jiujiang provenance from Jiangxi. Our preliminary conclusion is that the threshold of *C. paliurus* seedlings enduring salt was about 17.0 mol/L (1.0 g/L) NaCl.

The management of Na^+ movements within plants requires particular cell types in specific locations. Many reports have shown that solute transportation is different from water transport. Solutes such as Na^+, K^+, Ca^{2+}, Mg^{2+} and ABA move freely in the apoplast of roots and need to pass the Casparian bands of the endodermis to reach the apoplast of the xylem. Solute transport into the xylem is controlled by the plasma membrane of endodermal cells (Peterson et al. 1993; Blumwald et al. 2000; Kuhn et al.

2000; Steudle 2000; Chen et al. 2002). X-ray analysis of roots of *C. paliurus* seedlings under salt stress show that Na^+ is distributed mainly in the intercellular space of the endoderm while K^+ is concentrated in the cells and xylem to function in osmotic regulation. Generally, the variation of K^+ content in root tissues of *C. paliurus* seedlings contrasted with that of Na^+ content under salt stress, confirming that there is antagonism between Na^+ and K^+. Peng et al. (2004) reported in the case of antagonism between Na^+ and K^+ that the asymmetric distribution of K^+ and Na^+ changed relatively, even when channel block inhibitors were used, which indicated the barrier and filter functions of the endodermal cells. In the salt overly sensitive (SOS) mutants of *Arabidopsis*, the level of salt tolerance, as measured by root growth, is closely correlated with the K^+ content of the tissues (Zhu et al., 1998). Several other studies also suggested the importance of K^+ nutrition in salt tolerance (Peng et al., 2004). The results from our study indicated that *C. paliurus* seedlings are primarily sensitive to Na^+ excess, due to its adverse effects on K^+, Mg^{2+} and Ca^{2+} nutrition, cytosolic enzyme activities, photosynthesis and metabolism. Moreover, the results of our experiment also show that the relative content of Cl^- in stelar cells was high. This indicates that Cl^- can pass the Casparian bands by the pathways of apoplast and symplast, i.e., the damage to seedlings resulting from Cl^- toxicity was more serious than that from Na^+. To understand complex, whole-plant adaptations to salinity further, more information is required about cell-specific transport processes and the consequences of the manipulation of transporters in specific cell types, such as endodermal cells.

In response to the increasing concentrations of NaCl in the culture medium, plants have to cope with the lowering of external water potential. Consequently, the reduction of plant growth is usually attributed to osmotic stress (Song et al., 2005). Our results indicate salt stress significantly reduced seedling growth of *C. paliurus*, which agrees with those from studies on eucalyptus by Morabito et al. (1996). Based on the fact that increasing concentrations of NaCl induced a progressive decline both in survival and seedling growth (Fig. 2, Table 4), growth inhibition could be the result of ions, particularly sodium and chloride, likely to be absorbed into the plants to adjust their hydraulic potential in response to external stress. Unlike halophytes, glycophyte species seem unable to compartmentalize ions which become toxic from cellular metabolism (Greenway et al., 1980). *C. paliurus* did not depart from this behavior and salt tolerance of this species appears to be related to a low level of salt accumulation in leaves and stems compared to roots. The significant reductions in seedling growth at relatively mild external NaCl concentrations seem to indicate that *C. paliurus* is sensitive to salinity stress.

Acknowledgements We gratefully thank Prof. Yongqiang Mao of the Nanjing Institute of Geology and Paleontology, Chinese Academy of Sciences, for his help in measuring the relative ion content of the roots. Our study was supported by the National Natural Science Foundation of China (Grant No. 30371156) and the Project of Research for New High Technology in Jiangsu Province (No. BG2006314).

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