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RESEARCH ARTICLE

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# Effects of polyamines on $K^+$, $Na^+$ and $Cl^-$ content and distribution in different organs of cucumber (*Cucumis sativus* L.) seedlings under NaCl stress



**Abstract** Seedlings from the salt-sensitive cucumber cultivar Jinchun No. 2 and the salt-tolerant cucumber cultivar Changchun Mici were exposed for 8 days to 50 mmol/L NaCl in the absence or in the presence of exogenous foliar spraying PAs [putrescine (Put), spermidine (Spd), and spermine (Spm) 1 mmol/L] to compare the effects of different kinds of polyamines (PAs) on plant tolerance to salinity. This paper studied the effects of exogenous PAs on $K^+$, $Na^+$ and $Cl^-$ in different organs of cucumber seedlings. The results showed that $K^+$ content as well as the ratios of K/Na and Cl/Na decreased, while $Na^+$ and $Cl^-$ concentrations increased in salt-treated cucumber seedlings. The differences in $K^+$, $Na^+$ and $Cl^-$ content and the K/Na and Cl/Na ratios were greater for the salt sensitive cultivar Jinchun No. 2 than for the salt-tolerant cultivar Changchun Mici. Cucumber seedlings treated with exogenous polyamines and combined with salinity exhibited a higher level of $K^+$ accumulation and lower levels of $Na^+$ and $Cl^-$ accumulation compared with the seedlings treated only with salt stress. Among the three kinds of polyamines, Spd and Spm were more effective in inhibiting the accumulation of $Na^+$ and reduction of $K^+$. However, Put was more effective in reducing $Cl^-$ accumulation. Furthermore, all of the three kinds of exogenous polyamines could increase the ratio of K/Na, improving the absorption and transport selectivities of $K^+$ and $Na^+$ from stems to leaves for both cultivars. In conclusion, exogenous polyamines could alleviate salt damage to some extent and enhance the accumulation of biomass. Among the three kinds of polyamines, spermidine was most effective. Exogenous polyamines could improve tolerance of cucumber seedlings under salt stress by regulating the absorption and distribution of ions in different organs.

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## 1 Introduction

Soil salinity is one of the most crucial global problems in agricultural production. Today, almost 1 000 million $hm^2$ of land is affected by soil salinity worldwide, or 7% of the earth's total land area. Of the 1.5 billion $hm^2$ that is cultivated, about 5% (77 million $hm^2$) is affected by salt (Mark and Romola, 2003). In China, 27 million $hm^2$ of land belongs to saline-alkali soil, of which 7 million $hm^2$ is cultivated (Liu and Zhang, 1994). Increasing salinization has reached critical levels, with secondary soil salinization of protected cultivation now considered a major factor adversely affecting horticultural crops (Tong and Chen, 1991). Saline soil imposes both water deficit and ionic toxicity on plants, inducing deficiency in other nutrients (Munns, 2002). One of the primary plant responses to salinity is an increase of $Na^+$ and $Cl^-$ content, accompanied by a decrease in $Ca^{2+}$, $K^+$, $Mg^{2+}$ and $NO_3^-$ concentration in plant tissues (Khan et al., 2000), while excess $Na^+$ and $Cl^-$ in leaves may inhibit the growth and development of plants (Ashraf and Harris, 2004). Due to the physicochemical similarities between $Na^+$ and $K^+$, the competition of $Na^+$ at transport sites for $K^+$ entry into the symplast may result in $K^+$ deficiency. Hence, competition of cytoplasmic $Na^+$ for $K^+$ at binding sites may inhibit enzyme activities and metabolic processes that crucially depend on $K^+$ (Maathuis and Amtmann, 1999).

Polyamines (PAs), produced in nitrogen metabolism, form a kind of low-molecular-mass aliphatic amines that are ubiquitous in living organisms. The major forms are putrescine (Put), spermidine (Spd) and spermine (Spm). Polyamines have been associated with many aspects of plant growth and development. Since PAs were reported to be involved in stabilizing membrane, scavenging free radicals (Velikova et al., 1998), adjusting osmosis (Aziz et al., 1999), and regulating mineral nutrition (Tamai et al., 1999) and senescence

(Lahiri et al., 2004), their primary function has been considered to be adaptation response to protecting plants from various environmental stresses (Bouchereau et al., 1999). Additionally, the exogenous PAs could improve plant resistance to salt stress. The presoaked seed with Put could increase the germination percentage of bean (*Phaseolus vulgaris*) under salt stress (Zeid, 2004) and decrease $Na^+$ and $Cl^-$ accumulation in all organs of *Atropa belladonna* plant (Ali, 2000). Furthermore, the exogenous foliar spraying with Put might improve the growth of rice (*Oryza sativa* L.) under salt stress (Lutts et al., 1996). Most previous reports put particular emphasis on the effect of PAs on salt-tolerance of plants, although little is known about different physiological effects of PAs on plants subject to salt stress. The effects of PAs on plant under environmental stresses may actually depend on their kind (Kasukabe et al., 2004). To compare the effects of different kinds of exogenous polyamines on plant tolerance to salinity, elucidate the mechanism of exogenous polyamines in improving such tolerance, and develop suitable methods to mitigate the adverse effects of salinization on horticultural crops in protected cultivation, we investigated the effects of three kinds of exogenous polyamines, i.e. Put, Spd and Spm, on $K^+$, $Na^+$ and $Cl^-$ in different organs of two cucumber (*Cucumis sativus* L.) cultivar seedlings (with differential adaptation to salinity) after eight-day treatment with exogenous NaCl.

## 2 Materials and methods

### 2.1 Plant materials and treatments

The cucumber cultivars Changchun Mici and Jinchun No. 2 were used as plant materials in our study. Cucumber seedlings were grown in the glasshouse in Nanjing Agricultural University. Cucumber seeds were germinated in a culture tray lined with water moist filter paper and kept at 27°C for 24 h in the dark. Uniformly germinated seeds were then selected and sown in a plastic tray with quartz sand washed with tap water to remove any residual salts. Seedlings grew in natural daylight. Day temperatures ranged from 22°C to 28°C, and night temperatures were from 16°C to 18°C. When the cotyledons expanded, seedlings were watered with 1/2 strength Shanqi nutrient solution designed specially for cucumbers. When the second leaf fully expanded, seedlings of uniform size were transplanted into a water trough filled with one strength Shanqi nutrient solution. Solution pH was adjusted to $6.3 \pm 0.1$ and readjusted every two days. Solution was renewed every four days. To make the dissolved oxygen concentration in nutrient solution range from 6 to 8 mg/L, more air was compressed in the solution by an air pump working intermittently at 30 min/h. Three days later, when the cucumber seedlings were fully acclimated under control conditions, salt treatment began at 17:00 by adding exogenous NaCl into the nutrient solution, with the final NaCl concentration reaching 50 mmol/L. There were five treatments in this experiment, including:

(1) CK (control): Cucumber seedlings were cultivated in a nutrient solution without NaCl;

(2) NaCl: Cucumber seedlings were cultivated in a nutrient solution with 50 mmol/L NaCl;

(3) Put: Cucumber seedlings were cultivated in a nutrient solution with 50 mmol/L NaCl and leaves were sprayed with 1 mmol/L Put every day;

(4) Spd: Cucumber seedlings were cultivated in a nutrient solution with 50 mmol/L NaCl and sprayed on leaves with 1 mmol/L Spd every day;

(5) Spm: Cucumber seedlings were cultivated in a nutrient solution with 50 mmol/L NaCl and sprayed on leaves with 1 mmol/L Spm every day.

Foliar spraying in all of the treatments began at 18:00 every day. The solutions were all supplemented with 0.01% (v/v) Tween-20 as a detergent. Both CK and NaCl-treated seedlings were sprayed with 0.01% (v/v) Tween-20 solution.

Seedlings were harvested after eight days. For determination of the dry weight (DW), 15 plants per cultivar per treatment were collected. For analysis of $K^+$, $Na^+$ and $Cl^-$, cucumber seedlings were rinsed with deionized water to remove ions absorbed on the surface. The seedlings were separated into five parts from bottom to apex including the root (R), root-stem transition zone (G), stem (S), lower leaves (LL) and upper leaves (UL); deactivated enzyme at 105°C for 15 min; oven-dried at 70°C; ground into powder to pass through a 30-mesh screen; and stored for mineral analysis.

### 2.2 DW determination of individual plant

Cucumber seedlings were thoroughly washed with deionized water, deactivated enzyme at 105°C for 15 min, and dried at 70°C for 48 h in the blast oven to a constant weight.

### 2.3 Extraction and determination of $K^+$, $Na^+$ and $Cl^-$

The three kinds of ions of $K^+$, $Na^+$ and $Cl^-$ were extracted according to the second method described by Wang and Zhao (1995) with minor modifications. Dried samples of 50 mg were put into test tubes, where 20 mL of deionized water was added. After being fully oscillated, the test tubes were incubated in boiling-water for 1.5 h, cooled and filtered in volume-bottles. Concentrations of $K^+$ and $Na^+$ in the extracts were measured by atomic absorption spectrophotometry (Huipu 3500G). $Cl^-$ concentration in the extract was determined by a modified method of titration with $AgNO_3$ which had been described before (Lao, 1988), with the mixture of 4.2% (w/v) $K_2CrO_4$ and 0.7% $K_2Cr_2O_7$ as an indicator.

The selectivity ratio of $K^+$ to $Na^+$ ($S_{K,Na}$) was calculated as (Zheng et al., 2001)

Absorption $S_{K,Na} = ([K^+]/[Na^+])$ in root/($[K^+]/[Na^+]$) in nutrient solution;

Transporting $S_{K,Na} = ([K^+]/[Na^+])$ in pool organ/$([K^+]/[Na^+])$ in source organ.

The transporting selectivity ratio of $Cl^-$ to $Na^+$ ($S_{Cl,Na}$) was calculated as

$S_{Cl,Na} = ([Cl^-]/[Na^+])$ in pool organ/$([Cl^-]/[Na^+])$ in source organ.

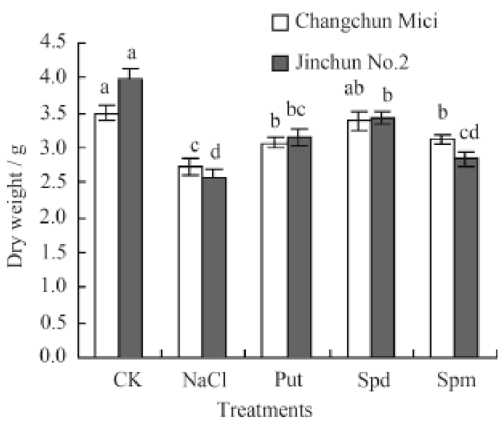
### 2.4 Statistical analysis

All statistical tests were performed using the computer software SAS. Means were compared using the Duncan's multiple-range test. Significant levels were reported as $P < 0.05$.

## 3 Results

### 3.1 Effects of exogenous PAs on individual plant DW of cucumber seedlings under NaCl stress

Compared with the control, the salt treatment significantly decreased individual plant DW of both cultivars (Fig. 1). DW was reduced by 22.12% and 35.24% of the control, respectively, for salt-resistant cultivar Changchun Mici and salt-sensitive cultivar Jinchun No. 2. Compared with the seedlings treated with NaCl only, DW of cucumber seedlings treated with exogenous PAs combined with salinity was obviously higher. Exogenous Put, Spd or Spm increased DW of Changchun Mici by 12.83%, 24.30% and 14.90%, whereas Jinchun No. 2 increased by 21.98%, 32.52% and 9.68%, respectively. Effects of three kinds of PAs were followed by the sequence of Spd > Put > Spm for Changchun Mici and Spd > Put > Spm for Jinchun No. 2. Exogenous Spd was the most effective among the three for both cultivars; the effects of PAs were thus most likely to depend on kinds and cultivars. Exogenous Spm significantly alleviated the detrimental effect of salt stress on Changchun Mici growth, but had little effect on Jinchun No. 2.




Notes: Different small letters represent significance at $P < 0.05$.

**Fig. 1** Effects of exogenous PAs on dry weight of cucumber seedlings under NaCl stress

### 3.2 Effects of exogenous PAs on contents, absorbing and transporting selectivity of $K^+$, $Na^+$ and $Cl^-$ in cucumber seedlings under NaCl stress

#### 3.2.1 Effects of exogenous PAs on contents of $K^+$, $Na^+$ and $Cl^-$

$K^+$ concentrations in NaCl-treated plants for both cultivars were significantly lower than those of the controls (Fig. 2). NaCl treatment reduced $K^+$ content for Changchun Mici by 47.12%, 13.98%, 23.60%, 29.17% and 10.94%, and by 54.32%, 31.26%, 15.91%, 32.37% and 18.40% for Jinchun No. 2 of the controls in R, G, S, LL and UL, respectively. The reduction in $K^+$ content over the controls was followed by the order of R > LL > S > C > UL and of R > LL > G > UL > S in different organs, respectively, for Changchun Mici and Jinchun No. 2. The reduction in $K^+$ content over the controls exhibited a higher extent in R and LL, and a lower level in UL. For salt-sensitive cultivar Jinchun No. 2, the reduction in $K^+$ content was higher than that for Changchun Mici. Compared with seedlings treated with NaCl only, $K^+$ content in all of organs of cucumber seedlings treated with exogenous PAs combined with salinity exhibited an obviously higher extent. Exogenous Put, Spd and Spm significantly increased $K^+$ content in S, LL and UL. Furthermore, Spd and Spm were more effective than Put. Exogenous PAs had a different effect on cultivars. For salt-tolerant cultivar Changchun Mici, the increment in $K^+$ content of PAs-treated seedlings was higher than that of the NaCl-treated seedlings in S and LL, and was lower in R, G and UL than that for Jinchun No. 2.

Compared with controls, salinity caused a significant rise of $Na^+$ concentration in R, G, S, LL and UL for both cultivars (Fig. 2). For salt-tolerant cultivar Changchun Mici, NaCl treatment increased $Na^+$ content by 9.83, 14.02, 13.81, 21.37 and 6.05 times compared with the controls in R, G, S, LL and UL, respectively. For salt-sensitive cultivar Jinchun No. 2, the increment was 13.33, 15.31, 15.20, 29.27 and 8.64 times that of the controls, respectively. For both cultivars, the difference in $Na^+$ content over the control was followed by the sequence of LL > S, G > R > UL in different organs. The increment in $Na^+$ content exhibited a lower extent in absorption organ R and photosynthesis organ UL, and $Na^+$ accumulated primarily in both G and S. $Na^+$ concentration was statistically higher for salt-sensitive cultivar Jinchun No.2. Compared with the seedlings treated with NaCl only, $Na^+$ content in seedlings treated with exogenous PAs combined with salinity were obviously lower in G, S, LL and UL of both cultivars. The reduction in $Na^+$ content over the NaCl treatment was followed by the sequence of LL and UL > G > S > R in different organs. The reduction in $Na^+$ content relative to the NaCl treatment was higher in leaves, but was lower in roots. The effects of three kinds of PAs were sequenced by Spd > Spm > Put.

Compared with the control, the change in $Cl^-$ content of NaCl-treated seedlings was minimal in R, but NaCl treatment induced a significant rise of $Cl^-$ concentration in G, S, LL

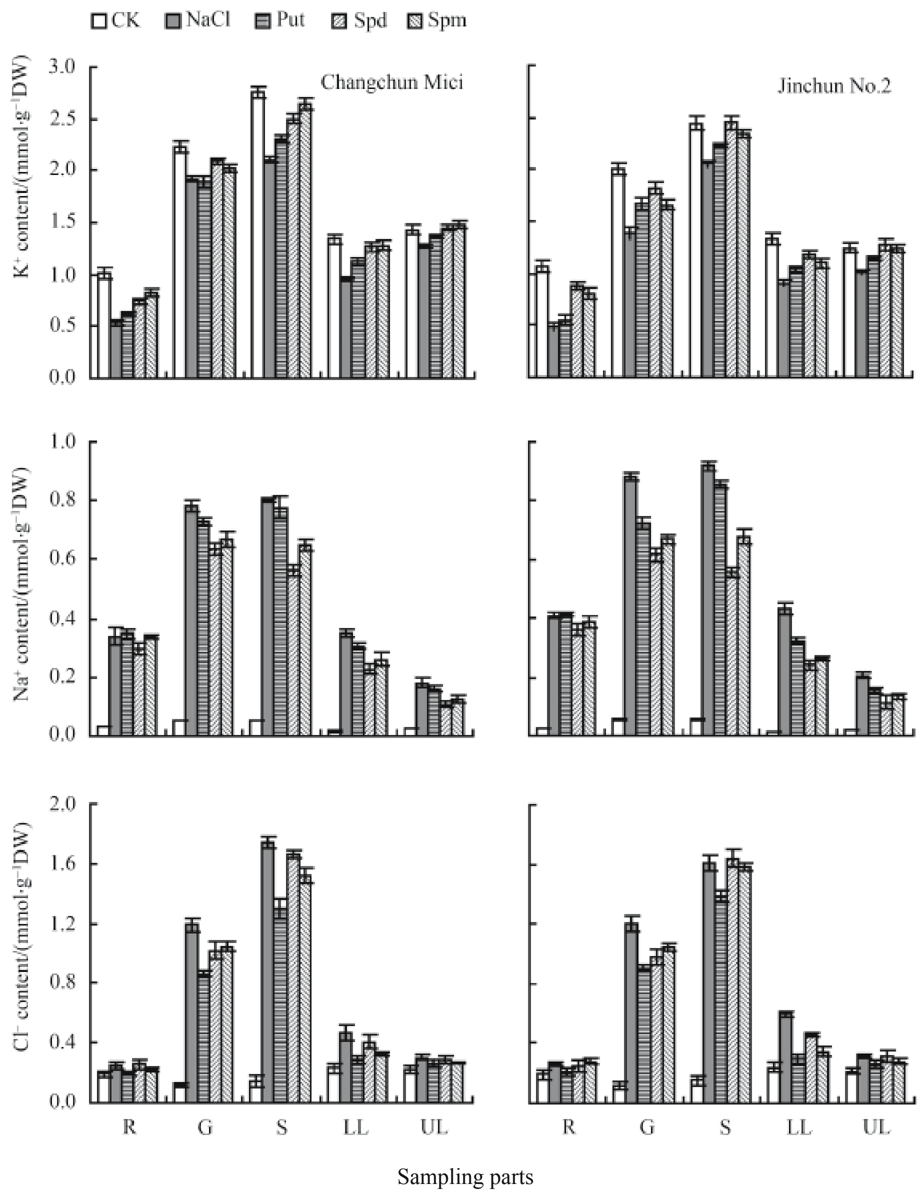



**Fig. 2** Effects of exogenous PAs on $K^+$, $Na^+$ and $Cl^-$ content in different organs of cucumber seedlings under NaCl stress

and UL for both cultivars (Fig. 2). For salt-tolerant cultivar Changchun Mici, $Cl^-$ content in NaCl-treated seedlings increased by 1.28, 10.16, 11.82, 2.03 and 1.36 times that of the control in R, G, S, LL and UL, respectively. For salt-sensitive cultivar Jinchun No. 2, $Cl^-$ content increased by 1.36, 9.75, 10.53, 2.53 and 1.48 times that of the control, respectively. For both cultivars, the increment in $Cl^-$ content over the controls was followed by the sequence of S > G > LL > UL > R in different organs. $Cl^-$ accumulated primarily in G, S and LL. Compared with the controls, $Cl^-$ accumulation was lower in R and UL. Compared with the seedlings treated with NaCl only, $Cl^-$ content of two cucumber cultivar seedlings treated with exogenous PAs combined with salinity exhibited an obviously lower extent in G, S and LL. The reduction in $Cl^-$ content over the NaCl treatment was higher in LL, with the effects of three kinds of PAs sequenced by Put > Spm > Spd.

### 3.2.2 Effects of exogenous PAs on K/Na ratio

Compared with the controls, NaCl treatment caused a large amplitude descent of K/Na ratios in all organs for both cultivars (Table 1). For salt-tolerant cultivar Changchun Mici, K/Na ratios reduced by 4.89%, 5.73%, 5.16%, 3.17% and 12.64% of the controls in R, G, S, LL and UL, respectively. For salt-sensitive cultivar Jinchun No. 2, K/Na ratios reduced by 4.22%, 7.72%, 5.19%, 2.24% and 8.47% of the controls, respectively. For both cultivars, K/Na ratios in different organs were followed by the sequence of UL > LL, S > G > R, increasing from bottom to apex, so that K/Na ratio was higher in UL. For salt-sensitive cultivar Jinchun No. 2, the reduction in K/Na ratio over the control was higher than that for Changchun Mici.

Exogenous PAs increased K/Na ratios in all organs in the seedlings of the two cultivars under NaCl stress (Table 1). For

**Table 1** Effects of exogenous PAs on K/Na ratio in different organs of cucumber seedlings under NaCl stress

| Treatment | Changchun Mici | | | | | Jinchun No. 2 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | R | G | S | LL | UL | R | G | S | LL | UL |
| CK | 32.18 | 42.82 | 50.91 | 85.31 | 55.85 | 37.31 | 37.19 | 43.03 | 93.67 | 57.92 |
| NaCl | 1.57 | 2.45 | 2.63 | 2.70 | 7.06 | 1.19 | 1.57 | 2.23 | 2.09 | 4.90 |
| Put | 1.77 | 2.59 | 2.97 | 3.65 | 8.57 | 1.33 | 2.30 | 2.62 | 3.22 | 7.33 |
| Spd | 2.49 | 3.30 | 4.42 | 5.56 | 13.29 | 2.46 | 2.96 | 4.40 | 4.88 | 11.07 |
| Spm | 2.45 | 3.02 | 4.07 | 4.91 | 11.55 | 2.08 | 2.48 | 3.47 | 4.18 | 9.07 |

both cultivars, the increment in K/Na ratios over the NaCl treatment was followed by the order of LL > UL > S, R, G in different organs. Exogenous PAs exhibited a higher effectivity to LL and UL. For salt-sensitive cultivar Jinchun No. 2, K/Na ratio relative to the NaCl treatment was greater than that for Changchun Mici, with the effects of three kinds of PAs sequenced by Spd > Spm > Put.

### 3.2.3 Effects of exogenous PAs on absorption and transporting $S_{K,Na}$

Compared with the control, NaCl treatment increased the absorption $S_{K,Na}$ of the root system for both cultivars Changchun Mici and Jinchun No. 2 (Table 2) by 4.17 and 2.72 times as much as that of the control, respectively. The increase in absorption $S_{K,Na}$ relative to the control was higher for Changchun Mici than for Jinchun No. 2. Exogenous PAs also obviously raised the absorption $S_{K,Na}$ in the seedlings of the two cultivars under salt stress. In comparison with the NaCl treatment, the combination of exogenous Put, Spd and Spm with salt stress caused an increase in absorption $S_{K,Na}$ by 12.71%, 58.45% and 55.84% for Changchun Mici, and 12.15%, 106.51% and 74.85% for Jinchun No. 2, respectively. Spd and Spm were more effective than Put.

NaCl treatment could increase the transporting $S_{K,Na}$ from R to the shoot for both cultivars (Table 2). For salt-resistant cultivar Changchun Mici, the increase in transporting $S_{K,Na}$ from R to the shoot relative to the control was primarily caused by a rise of transporting $S_{K,Na}$ from S to leaves. For salt-sensitive cultivar Jinchun No. 2, the rise of transporting $S_{K,Na}$ was from R to S. Moreover, NaCl treatment could induce a rise of transporting $S_{K,Na}$ from S to UL in both cultivars compared with the control. The increase in transporting $S_{K,Na}$ from S to UL relative to the control was 142.96% and 70.25%, respectively, for Changchun Mici and Jinchun No. 2. Thus, the transporting $S_{K,Na}$ of Changchun Mici from S to UL was higher than that of the control, indicating that $K^+$ was preferentially supplied to UL by increasing the transporting selectivity of $K^+$ to $Na^+$ to UL. Transporting $S_{K,Na}$ from S to leaves was positively correlated with tolerance to salt stress.

For the two cultivars, exogenous PAs had different effects on transporting $S_{K,Na}$ from R to S under salt stress. For salt-tolerant cultivar Changchun Mici, exogenous PAs caused a descent of transporting $S_{K,Na}$ from R to G compared with NaCl treatment only, but a rise of $S_{K,Na}$ from G to S (Table 2). As a result, exogenous PAs made more $Na^+$ accumulate in G in Changchun Mici seedlings. Among three kinds of PAs, Spd and Spm were more effective than Put. However, exogenous PAs had no effect on Jinchun No. 2. Compared with NaCl treatment only, exogenous PAs increased transporting $S_{K,Na}$ from R to S in Changchun Mici and could cause a rise of transporting $S_{K,Na}$ from S to leaves for both cultivars. In conclusion, exogenous PAs could affect the allocation and translocation of $K^+$ and $Na^+$ among different organs in salt-stressed cucumber seedlings.

**Table 2** Effects of exogenous PAs on root absorption $S_{K,Na}$ and transporting $S_{K,Na}$ between different organs in cucumber seedlings under NaCl stress

| Cultivar | Treatment | Absorption $S_{K,Na}$ | Transporting $S_{K,Na}$ from R to shoot | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | R to S | | | S to leaves | | | R to shoot |
| | | | R to G | G to S | R to S | S to LL | S to UL | S to leaves | |
| Changchun Mici | CK | 3.14 | 1.33 | 1.19 | 1.55 | 1.71 | 1.12 | 1.28 | 1.74 |
| | NaCl | 13.09 | 1.56 | 1.07 | 1.65 | 1.04 | 2.72 | 1.65 | 2.03 |
| | Put | 14.76 | 1.46 | 1.14 | 1.63 | 1.27 | 2.97 | 1.77 | 2.06 |
| | Spd | 20.75 | 1.33 | 1.34 | 1.70 | 1.32 | 3.15 | 1.88 | 2.16 |
| | Spm | 20.41 | 1.23 | 1.35 | 1.56 | 1.29 | 3.03 | 1.85 | 1.99 |
| Jinchun No.2 | CK | 3.64 | 1.00 | 1.16 | 1.13 | 2.21 | 1.37 | 1.63 | 1.38 |
| | NaCl | 9.92 | 1.32 | 1.42 | 1.77 | 0.99 | 2.33 | 1.54 | 2.11 |
| | Put | 11.12 | 1.73 | 1.14 | 1.93 | 1.25 | 2.85 | 1.88 | 2.44 |
| | Spd | 20.48 | 1.2 | 1.49 | 1.69 | 1.18 | 2.67 | 1.72 | 2.08 |
| | Spm | 17.34 | 1.19 | 1.40 | 1.57 | 1.28 | 2.78 | 1.93 | 1.98 |

### 3.2.4 Effects of exogenous PAs on Cl/Na ratio and transporting $S_{Cl,Na}$

NaCl treatment could obviously reduce Cl/Na ratios in all organs of the seedlings of the two cucumber cultivars (Table 3). For salt-tolerant cultivar Changchun Mici, Cl/Na ratios decreased by 88.14%, 32.39%, 20.16%, 90.93% and 80.65% of the controls in R, G, S, LL and UL, respectively. For salt-sensitive cultivar Jinchun No. 2, Cl/Na ratios decreased by 90.50%, 40.19%, 34.97%, 91.65% and 84.64%, respectively. For both cultivars, the reduction in Cl/Na ratios over the controls was followed by the order of LL, R > UL > G > S in different organs, suggesting that the reduction in Cl/Na ratios relative to the controls was higher in both R and leaves. Compared with NaCl treatment only, three kinds of exogenous PAs had different effects on Cl/Na ratios. Exogenous Put reduced Cl/Na ratios in all organs, but exogenous Spd increased Cl/Na ratios under salt stress. Exogenous Spm could also cause a rise of Cl/Na ratios in S and UL under salt stress.

For both cultivars, NaCl treatment resulted in a rise of transporting $S_{Cl,Na}$ from R to shoot (Table 4). The increase in transporting $S_{Cl,Na}$ from R to S was mainly due to the increase in transporting $S_{Cl,Na}$ from R to G. The transporting $S_{Cl,Na}$ from S to both LL and UL could decrease under salt stress, which suggested that accumulation of $Na^+$ in the leaves relative to the control was greater than that for $Cl^-$. Therefore, cucumber seedlings appeared more sensitive to excess $Na^+$ than excess $Cl^-$.

Compared with NaCl treatment only, exogenous PAs had no obvious effects on the transporting $S_{Cl,Na}$, except that transporting $S_{Cl,Na}$ from S to UL exhibited a minor rise (Table 4). Exogenous Put could also slightly affect the transporting $S_{Cl,Na}$ from R to S; the three kinds of PAs could therefore improve the transporting $S_{Cl,Na}$ from R to shoot due to a rise of transporting $S_{Cl,Na}$ both from R to G and from G to S. These results indicated that exogenous Spd was more effective.

## 4 Discussion

Ion homeostasis at both whole plants and cell levels is vital to the growth and development of salt-stressed plants. $K^+$ is essential to all plant life, and in most terrestrial plants, it is the major cationic inorganic nutrient. Cellular $K^+$ plays a key role in several physiological processes such as regulating ion balance, regulating membrane permeability, keeping cellular turgor, influencing protein synthesis and enhancing photosynthesis. Excess $Cl^-$ may interfere with anionic sites involved in the binding of RNA and anionic metabolites such as bicarbonate, carboxylates and sugar-phosphates (Serrano et al., 1999). One of the primary plant responses to salinity is an influx of $Na^+$ and a decrease of $K^+$ concentration in plant tissues. Evidence has been advanced that one of mechanisms by which plants tolerate salt stress depends on their capacity to maintain a lower $Na^+$ and $Cl^-$ influx and a higher $K^+$ concentration (Shannon and Grieve, 1999). The degree to

**Table 3** Effects of exogenous PAs on Cl/Na ratio in different organs of cucumber seedlings under NaCl stress

| Treatment | Changchun Mici | | | | | Jinchun No. 2 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | R | G | S | LL | UL | R | G | S | LL | UL |
| CK | 6.13 | 2.25 | 2.72 | 14.66 | 8.64 | 6.75 | 2.28 | 2.68 | 16.57 | 9.89 |
| NaCl | 0.73 | 1.52 | 2.17 | 1.33 | 1.67 | 0.64 | 1.36 | 1.74 | 1.38 | 1.52 |
| Put | 0.57 | 1.18 | 1.67 | 0.94 | 1.62 | 0.51 | 1.25 | 1.63 | 0.91 | 1.66 |
| Spd | 0.86 | 1.60 | 2.94 | 1.77 | 2.68 | 0.71 | 1.59 | 2.94 | 1.90 | 2.72 |
| Spm | 0.66 | 1.56 | 2.34 | 1.23 | 2.07 | 0.72 | 1.57 | 2.34 | 1.32 | 2.04 |

**Table 4** Effects of exogenous PAs on transporting $S_{Cl,Na}$ between different organs of cucumber seedlings under NaCl stress

| Cultivar | Treatment | From R to shoot | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | R to S | | | S to leaves | | | R to shoot |
| | | R to G | G to S | R to S | S to LL | S to UL | S to leaves | |
| Changchun Mici | CK | 0.37 | 1.21 | 0.43 | 5.52 | 3.25 | 3.86 | 0.98 |
| | NaCl | 2.09 | 1.43 | 2.82 | 0.65 | 0.82 | 0.71 | 2.54 |
| | Put | 2.09 | 1.41 | 2.76 | 0.60 | 1.04 | 0.73 | 2.50 |
| | Spd | 1.87 | 1.83 | 3.14 | 0.66 | 0.99 | 0.76 | 2.91 |
| | Spm | 2.37 | 1.50 | 3.27 | 0.57 | 0.96 | 0.70 | 2.95 |
| Jinchun No.2 | CK | 0.34 | 1.18 | 0.39 | 6.29 | 3.76 | 4.54 | 0.86 |
| | NaCl | 2.13 | 1.28 | 2.61 | 0.83 | 0.91 | 0.86 | 2.48 |
| | Put | 2.47 | 1.31 | 3.10 | 0.58 | 1.06 | 0.77 | 2.88 |
| | Spd | 2.26 | 1.85 | 3.83 | 0.70 | 1.01 | 0.81 | 3.59 |
| | Spm | 2.17 | 1.49 | 3.01 | 0.61 | 0.94 | 0.75 | 2.79 |

which glycophytes tolerate salt stress is correlated with their capacity to prevent the absorption and transport to shoot of toxic ion (Greenway and Munns, 1980). The basic strategy includes restricting $Na^+$ influx to shoot and compartmentalizing excess toxic ions in the root system, stem bottom (Ashraf and O'Leary, 1995) and fully expanded leaves (Loupassaki et al., 2002). When the accumulation of toxic ions in upper leaves are restricted closely, the more $K^+$ can be supplied for new growing tissues (Khatun and Flowers, 1995). In the present experiment, the results showed that NaCl treatment resulted in an increase in $Na^+$ and $Cl^-$ concentration and a reduction in $K^+$ concentration (Fig. 2). $Na^+$ and $Cl^-$ accumulated primarily in lower leaves and stems in salt-stressed cucumber seedlings, whereas $Na^+$ and $Cl^-$ content in root systems and upper leaves instead of the other organs were lower than those of the controls, and the reduction in $K^+$ concentration in upper leaves was the lowest in all organs. NaCl treatment increased the absorption selectivity and transporting selectivity of $K^+$ to $Na^+$ from root to shoot and from stem to upper leaves, and decreased transporting selectivity of $K^+$ to $Na^+$ from the stem to lower leaves. As a result, K/Na ratios in cucumber seedlings increased from bottom to apex (Tables 1 and 2). The higher K/Na ratio in upper leaves compared with the other organs could supply enough $K^+$ for photosynthesis. Therefore, cucumber seedlings under salt stress could compartmentalize the toxic ions at the organ level, which was one of the adaptive mechanisms by which cucumber seedlings responded to salt stress. However, for many plants, $Na^+$ is viewed as the primary cause of ion-specific damage (Mark and Romola, 2003). NaCl treatment could decrease Cl/Na ratios in all organs in cucumber seedlings compared with the control, especially in root and leaves (Table 3). Furthermore, NaCl treatment could increase the transporting selectivity of $Cl^-$ to $Na^+$ from root to stem, but decrease transporting selectivity of $Cl^-$ to $Na^+$ from stem to leaves. This indicates that the compartmentalization of $Cl^-$ in the stem was higher than that of $Na^+$ (Table 4), whose concentration in leaves relative to the control was greater than that of $Cl^-$, which might be an important cause of ion-specific damage in salt-stressed cucumber seedlings.

Polyamines are basic molecules which are positively charged at physiological pH. They are shown to bind strongly with negatively charged nucleic acids and acidic phospholipids on plasma membrane. These ionic interactions are important to plants under salt stress in preventing degradation of biological macromolecules and alleviating membrane damage (Basra et al., 1994). Recently, evidence indicated that PAs played an important role in plant responses to environmental stress. Exogenous Put and Spd increased the activities of root tonoplast $H^+$-ATPase, $H^+$-PPiase and $Na^+/H^+$ transporter in barley seedlings under salt stress (Zhao and Qin, 2004). Furthermore, exogenous Spd increased $H^+$-ATPase activity on plasma membrane in rice under salt stress (Roy et al., 2005). Exogenous PAs could protect the integrity of plasma membrane and tonoplast under salinity, and regulate the absorption and allocation of ions in plant cells. Therefore, PAs could improve plant tolerance to salt stress. In our experiment, exogenous Put, Spd and Spm inhibited the reduction of $K^+$ concentration in all organs and the increase of $Na^+$ and $Cl^-$ concentration in stems and leaves, resulting in higher K/Na ratios in all organs compared with NaCl treatment, especially in leaves (Fig. 2, Table 1). Polyamines caused a rise of absorption selectivity of $K^+$ to $Na^+$ of the root system and transport selectivity from root to shoot, and increased transport selectivity of $Cl^-$ to $Na^+$ from root to stem (Tables 2 and 4). This is consistent with other reports that exogenous PAs could regulate ion homeostasis and absorption and translocation of toxic ions such as exogenous Put in rice (Krishnamurthy, 1991), exogenous Put and Spd in spring wheat (*Triticum aestivul* L.) (Iqbal and Ashraf, 2005) and exogenous Spd in maize (Jiang et al., 2000). Ndayiragije and Lutts (2006) reported that in rice, the regulating effect of exogenous Put on ion balance was due to the accumulated endogenous Put rather than competition between cationic PAs and $Na^+$ at the site of absorption. Exogenous PAs induced the change in endogenous PAs content, and influenced $Na^+$ partition and $K^+$ versus $Na^+$ discrimination at the sites of xylem loading. In brief, application of exogenous PAs resulted in the alteration of endogenous PAs content, then indirectly regulated osmosis and ion homeostasis by improving the stabilization of cell membrane structure. Hence, exogenous PAs could improve the salinity tolerance of plants.

In conclusion, salt stress can result in an increase in $Na^+$ and $Cl^-$ concentration concomitantly with a decrease in $K^+$ and K/Na ratio. Exogenous PAs may have a positive impact on monovalent cation discrimination in the process of absorption and transport, regulating the absorption and translocation of toxic ions and $K^+$, and alleviating the deleterious effect of salt stress on cucumber seedlings. Exogenous Spd proved to be the most effective in PAs.

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