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# Uptake and distribution of N, P and heavy metals in three dominant salt marsh macrophytes from Yangtze River estuary, China

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## Abstract

We examined the variation in aboveground biomass accumulation and tissue concentrations of nitrogen (N), phosphorus (P), copper (Cu), zinc (Zn) and lead (Pb) in *Phragmites australis* (common reed), *Spartina alterniflora* (salt cordgrass), and *Scripus mariqueter* throughout the growing season (April-October 2005), in order to determine the differences in net element accumulation and distribution between the three salt marsh macrophytes in the Yangtze River estuary, China. The aboveground biomass was significantly greater in the plots of *S. alterniflora* than in the plots of *P. australis* and *S. mariqueter* throughout the growing season ( $P < 0.05$ ). In August, the peak aboveground biomass was  $1246 \pm 89$  gDW/m<sup>2</sup>,  $2759 \pm 250$  gDW/m<sup>2</sup> and  $548 \pm 54$  gDW/m<sup>2</sup> for *P. australis*, *S. alterniflora* and *S. mariqueter*, respectively. The concentrations of nutrients and heavy metals in plant tissues showed similar seasonal patterns. There was a steady decline in element concentrations of the aboveground tissues from April to October. Relative element concentrations in aboveground tissues were at a peak during the spring sampling intervals with minimum levels during the fall. But the concentrations of total nitrogen and total phosphorus in the belowground tissues were relatively constant throughout growing season. Generally, trace metal concentrations in the aboveground tissues of *S. mariqueter* was the highest throughout the growing season, and the metal concentrations of *S. alterniflora* tissues (aboveground and belowground) were greater than those of *P. australis*. Furthermore, the aboveground pools of nutrients and metals were consistently greater for *S. alterniflora* than for *P. australis* and *S. mariqueter*, which suggested that the rapid replacement of native *P. australis* and *S. mariqueter* with invasive *S. alterniflora* would significantly improve the magnitude of nutrient cycling and bioavailability of trace metals in the salt marsh and maybe transport more toxic metals into the water column and the detrital food web in the estuary.

**Keywords:** Yangtze River estuary; Nutrients; Metals; Salt marshes; *Phragmites*; *Spartina*

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## 1. Introduction

Macrophytes play important roles in the biogeochemistry of salt marsh ecosystems through their active and passive circulation of elements (Weis & Weis, 2004; Weis, Windham, Burke, & Weis, 2002). Many estuarine salt marshes near urbanized areas receive substantial quantities of anthropogenic materials, including nutrients and metals. When marsh sediment accumulates high concentrations of nutrients and metals, macrophyte roots can absorb these materials and translocate them to aboveground biomass, which may lead to the increase of nutrient and metal bioavailability in the salt marsh (Windham, Weis, & Weis, 2003; Windham, 1999). Macrophytes in salt marshes are widely believed to decrease the rate of tidal flow, cause suspended particulate matter to sediment, enhance the retention rates of nutrients and metals from overlying water, and reduce the import of nutrients and metals to estuarine waters (Cacador, Vale, & Catarino, 2000; Daehler & Strong, 1996; Windham et al., 2003; Windham, 1999). Therefore, salt marshes are often considered efficient sinks for nutrients and metals; they can ameliorate the environmental quality in coastal ecosystem (Romero, Comin, & Garcia, 1999; Coelho, Flindt, & Jensen, 2004; Zhang, Yu, Hutchinson, Xu, Chen, & Gao, 2001).

Salt marsh macrophytes have high primary productivity and assimilative capacity for nitrogen (N) and Phosphorus (P) (Romero et al., 1999; Teal & Howes, 2002). With denitrification as an exception, plant uptake is thought to be the important mechanism for salt marsh ecosystems to remove inorganic nutrients (N and P) from overlying water (White, 1994; Teal & Howes, 2002; Rozema, Leendertse, Bakker, & Wijnen, 2002). With increasing N supply, salt marshes can show greater primary productivity and withstand large additions of nutrients without severe damage (Adcock & Ganf, 1994; Brix & Schierup, 1989; White, 1994). Previous studies focusing on P dynamics show that mud flats colonised by *Spartina* marshes have higher adsorption capacity, and—through primary production activity—these macrophytes were able to control phosphate efflux from the sediment (Teal & Howes, 2002; Coelho et al., 2004). Therefore, salt marsh plant activities significantly influence the biogeochemistry of nutrients and increase the retention rate of N and P in salt marshes.

Metal dynamics in salt marshes may be influenced by the composition of plant communities (Williams, 2002; Windham et al., 2003). Recent investigations demonstrate that concentrations of metals in plant tissues are highly variable between species (Fitzgerald, Caffrey, Nesaratnam, & McLoughlin, 2003; Ye, Baker, Wong, & Willis, 1997). Differences in uptake rates and allocation

patterns between species can generate different rates of metal retention in salt marsh vegetation and perhaps further influence the biogeochemistry of salt marshes (Windham et al., 2003; Burke, Weis, & Weis, 2000; Weis & Weis, 2004; Ornes, Sajwan, Loganathan, & Chetty, 1998).

In many salt marshes on the Atlantic coast of North America, *Phragmites australis* has been invading the low marsh zone dominated by *Spartina alterniflora*. Several studies demonstrate that *P. australis* expansion could alter edaphic conditions (Windham & Lathrop, 1999; Bart & Hartman, 2000), accelerate nutrient cycling (Windham, 1999; Meyerson, Saltonstall, Windham, Kiviat, & Findlay, 2000), and increase metal bioavailability (Weis & Weis, 2004; Weis et al., 2002; Windham et al., 2003). The genus *Spartina* is native to the Atlantic and Gulf Coasts of North America, but it has been introduced to many coastal and estuarine regions of the world (Daehler & Strong, 1996; Chen, Li, Zhong, & Chen, 2004). On the Pacific Coast of North America, four introduced cordgrass species have seriously invaded several estuaries. These invading *Spartina* have outcompeted the native salt marsh plant, reduced open mud feeding habitat for shorebirds and fish, caused suspended sediment to precipitate, and altered the trophic function of salt marsh ecosystems (Daehler & Strong, 1996; Levin, Neira, & Grosholz, 2006). In China, *S. alterniflora* was intentionally introduced from North America in 1979 for erosion control, soil melioration, and dike protection. Today, it is spreading along the east coast of China from Tianjin to Baihai in Guangxi.. Small patches of *S. alterniflora* were first found at Dongtan wetland on Chongming Island of the Yangtze River estuary in 1995, which are believed to have invaded the mudflats through seed dispersal from Qidong by water flow. More importantly, *S. alterniflora* was transplanted to Dongtan and Jiuduansha wetlands for rapid sediment accretion in the late 1990s when many salt marshes were regularly reclaimed in the Yangtze River estuary. Since then, *S. alterniflora* has rapidly expanded in salt marshes and gradually replaced the native plants (*P. australis* and *Scirpus mariqueter*). *S. alterniflora* is now a dominant salt marsh macrophyte in the Yangtze River estuary (Chen, Ma, & Li, 2003).

The shift in plant community toward *S. alterniflora* monoculture may lead to different patterns of nutrient and metal cycling in salt marsh ecosystems. Few studies have examined the impact of the *S. alterniflora* invasion on the element cycling of the salt marsh ecosystem in the Yangtze River estuary. Our objective is to explore the influence of *S. alterniflora* invasion on nutrient and metal cycling in the salt marsh ecosystem in the Yangtze River estuary. We describe herein the uptake and distribution of N, P and heavy metals (Cu, Zn and Pb) in the plant tissues of *P. australis*, *S. alterniflora* and *S.*

*mariqueter* over a growing season (April-October, 2005).

## 2. Materials and methods

### 2.1 Study site

The Dongtan wetland at Chongming Island, Shanghai City, is in the Yangtze River estuary of China (Figure 1). In this wetland, *P. australis*, *S. alterniflora* and *S. mariqueter* were the three salt marsh macrophytes. Each plant can form a dense monoculture in the proper marsh zone. Monocultures of *P. australis* are often found around the high marsh zone, while *S. mariqueter*, a pioneer plant, dominates low marsh zones. In the late 1990s, *S. alterniflora* was transplanted to the middle marsh zone and is now quickly expanding toward the higher and lower marsh zones. At this study site, concentrations of heavy metals in the sediment are moderate, compared with other salt marshes in Asia and Europe (Zhang et al., 2001). Average metal concentrations in the sediment during the growing season of 2005 were Cu-42±11mg/kg; Zn-122±23 mg/kg; Pb-37±10 mg/kg. (Quan, unpublished data).

### 2.2 Field sampling and analysis methodology

We visited the study site every other month throughout the growing season (April, June, August, and October of 2005). During each field sampling, replicate 1m<sup>2</sup> plots (n=6) were staked in each vegetation type (*P. australis*, *S. alterniflora* and *S. mariqueter*) for collection of standing live aboveground biomass. Six soil cores (diameter 15cm and depth 30cm) were sampled in each vegetation type. All cores were washed over a 1mm mesh screen. The retained live roots and rhizomes (white) were collected for analysis. All the samples were sent immediately to a laboratory for processing.

All plant samples were washed thoroughly with tap water and then cleaned with deionised water. Samples were dried at 60°C to constant weight for approximately 48h. The dry plant samples were then ground to powder with a mortar and pestle. Sub samples for each sampled plot and tissue type were analyzed for elemental concentrations (TN, TP, Cu, Zn and Pb). N concentration of the plant tissues was determined by the Kjeldahl method. 0.2g sub samples of plant tissues were digested with HNO<sub>3</sub>-HCl (3:1 v/v) at 200°C, and then P in the solution was determined by the spectrophotometer method. Ten millilitres of HNO<sub>3</sub>/HClO<sub>4</sub> (5:1 v/v) were added to 0.2g of dry plant sample. The mixture was heated slowly to approximately 100°C, until the formation of nitrous fumes stopped. Then the mixture was boiled to approximately 200°C, until most of the liquid evaporated. The remaining extract

was diluted with deionised water to 25ml. The metal concentrations (Cu, Zn and Pb) in the solution were determined by the flame atomic absorption spectrophotometer (3500G, HP inc). All the chemical reagents were GR grade. Inter-laboratory standard reference material and method blanks were digested and analyzed for Cu, Zn and Pb with the same method. The concentration on average deviated 3.24% (Cu), 6.71% (Zn) and 5.10% (Pb) from certified average values. The aboveground biomasses of the three salt marsh macrophytes were calculated on a dry weight basis ( $\text{g/m}^2$ ). The elemental concentration and biomass of aboveground tissues were multiplied to yield aboveground element pools.

### 2.3 Data analysis

All data was log-transformed to meet the parametric assumptions prior to statistical analysis. One-way analysis of variances (ANOVA) was carried out to compare the aboveground biomass and elemental concentrations among the three plant species using STATISTICA 6.0 (StatSoft Inc, 2001). Duncan's test method was used to test the significant difference between the three salt marsh macrophytes. A set of linear regressions were performed between the elemental concentrations in plant tissue and time for the three species. Each pair of regressions was compared with covariance analyses (ANCOVA). Differences were considered significant when the probabilities for non-significance were smaller than 0.05.

## 3. Results

### 3.1 Aboveground biomass

*S. mariqueter* was not emergent aboveground in the first field sampling (April of 2005). The aboveground biomasses of the three species showed the same seasonal variation pattern. From April of 2005, the aboveground biomasses of the three species increased gradually. By August of 2005, the macrophytes reached maximum aboveground biomasses (Figure 2). One-way ANOVA found that the aboveground biomass was significantly greater in the plots of *S. alterniflora* than in the plots of the two native plants (*P. australis* and *S. mariqueter*) (Figure 2,  $P < 0.05$ ). The aboveground biomasses for *S. alterniflora* were 120-186% greater than *P. australis* and 300-600% greater than *S. mariqueter*. The aboveground biomass in the plots of *P. australis* had no significant difference from the the plots of *S. mariqueter*, except in August. The results showed that *S. alterniflora* had greater aboveground biomass than native plant across the whole growing season.

### 3.2 Elemental Concentration

Seasonal patterns of total N (TN) and total P (TP) concentration in plant tissue were relatively consistent among the three species (Figure 3). From April to October 2005, concentrations of TN and TP in the aboveground tissues showed a gradually decreasing trend, whereas the concentrations of TN and TP in the belowground tissues were relatively constant over the growing season (Figure 3). A linear regression between nutrient concentrations in plant tissues and time was established for the three species (Table 1). A significantly negative regression relationship was found between aboveground nutrient concentrations and time for *P. australis* and *S. alterniflora* (Table 1,  $P < 0.01$ ). The regression between belowground nutrient concentrations and time was not significant for the three species. Analysis of covariance revealed significant differences in the regression between belowground TN concentration and time for the three species (Table 1, ANCOVA,  $P < 0.01$ ). The differences in the regression between aboveground TN vs. time, aboveground TP vs. time, and belowground TP vs. time were not significant for the three species (Table 1, ANCOVA,  $P > 0.05$ ).

For the three plant species, the concentrations of TN and TP were obviously greater in the aboveground plant tissues than the belowground tissues (Figure 3). TN concentrations in the aboveground tissues of *P. australis* and *S. alterniflora* were similar throughout the growing season and lower than *S. maritima*. TN concentrations of aboveground tissues were significantly greater in *S. maritima* than in both *P. australis* and *S. alterniflora*, especially in October (Figure 3,  $P < 0.05$ ). For the belowground tissues, from April to August, TN concentrations were similar among the three species. In October, however, TN concentrations were significantly greater in *P. australis* than in both *S. alterniflora* and *S. maritima* (Figure 3,  $P < 0.05$ ).

In the aboveground tissues, TP concentration in *P. australis* was the lowest of the three species over the growing season (Figure 3). In June, TP concentration of the aboveground tissue was significantly lower in *P. australis* than in *S. alterniflora* and *S. maritima* (Figure 3,  $P < 0.05$ ). By October, similar to TN results, TP concentration of the aboveground tissue was significantly greater in *S. maritima* than in *P. australis* and *S. alterniflora* (Figure 3,  $P < 0.05$ ). But in the belowground tissues, TP concentration in *P. australis* and *S. alterniflora* were similar and consistently lower than *S. maritima*. In August and October, TP concentrations in *S. maritima* were significantly greater than *P. australis* and *S. alterniflora* (Figure 3,  $P < 0.05$ ).

Metal concentrations in plant tissues over the growing season were shown in Figure 4. The aboveground and belowground tissues in the three species contained different levels of Cu, Zn, and Pb.



Metal concentrations were greater in belowground tissues than aboveground tissues. But seasonal variation patterns of metal concentrations in plant tissues were similar between the three species. Early in the growing season (April), relatively higher metal concentrations accumulated in the tissues of the three macrophytes (Figure 4). From April to October, metal concentrations in the plant tissues showed a gradual decreasing trend. At the same time, a negative regression relationship between metal concentrations in plant tissues and time was established for the three species (Table 2). Analysis of covariance found no significant differences in the regression between aboveground Cu vs. time and belowground Cu and Zn vs. time. Analysis of covariance, however, revealed significant differences in the regression between aboveground Zn and Pb vs. time (Table 2, ANCOVA,  $P<0.05$ ) and belowground Pb vs. time (Table 2, ANCOVA,  $P<0.01$ ) for the three species.

Among the metals analyzed, Zn was the most abundant in all parts of the plants. Cu concentrations in the aboveground tissues of *S. mariqueter* were the highest among the three species, except in April. In October, Cu and Pb concentrations in the aboveground tissues were significantly greater in *S. mariqueter* than *P. australis* and *S. alterniflora* (Figure 4,  $P<0.05$ ). From June to October, relatively low metal concentrations accumulated in the aboveground tissues of *P. australis*. In June, Cu concentrations in the aboveground tissues were significantly lower in *P. australis* than *S. alterniflora* and *S. mariqueter* (Figure 4,  $P<0.05$ ). The same resulted for Zn in August.

Belowground concentrations of the three metals were more variable between the species throughout the growing season. From April to August, Cu and Zn concentrations in *S. alterniflora* were the highest of the three species. Cu and Zn concentrations in the belowground tissue were significantly greater for *S. alterniflora* in April and August than for both native plants (Figure 4,  $P<0.05$ ). Similar to the aboveground tissues, the metal concentrations in belowground tissues of *P. australis* were the lowest among the three plants in October. For Zn and Pb, the belowground concentrations were significantly lower in *P. australis* and *S. alterniflora* than *S. mariqueter* in October (Figure 4,  $P<0.05$ ).

### 3.3 Aboveground pools

The aboveground pools of given elements were calculated by combining elemental concentration with aboveground biomass. Aboveground elemental pools showed clear seasonal variation patterns throughout the growing season. Seasonal patterns of both nutrients and metals had the minima in April and the maxima in August at the peak of the growing season (Figure 5, Figure 6).

Throughout the growing season, the aboveground pools of both N and P were significantly greater in *S. alterniflora* than *P. australis* and *S. maritima* (Figure 5,  $P < 0.05$ ), owing to greater aboveground biomass in the plots of *S. alterniflora* (Figure 5). The aboveground N pools were 23-192% greater in *S. alterniflora* and 190-265% greater in *S. maritima* than *P. australis*. The aboveground TP pools of *S. alterniflora* were approximately 3.1 and 3.4 times higher than *P. australis* and *S. maritima*, respectively. Except for TN in August, the aboveground TN and TP pools showed no significant difference between the two native species.

Similar to the aboveground pools of N and P, the aboveground pools of metals (Cu, Zn and Pb) showed the same seasonal variation pattern (Figure 6). In August, all the plants reached maximum pools of metals. The aboveground pools of metals were significantly greater in *S. alterniflora* than in both *P. australis* and *S. maritima* (Figure 6,  $P < 0.05$ ). The aboveground metal pools showed no significant difference between the two native plants throughout the growing season, especially for Cu in August. The results showed that exotic plant *S. alterniflora* could translocate significantly more nutrients and metals to the aboveground tissue than the two native plants.

#### 4. Discussion

Our measurements of peak aboveground biomass were not equal to the annual net production of the aboveground parts due to significant biomass losses from shoot mortality and leaf shedding. Shoot growth started in March, at the expense of rhizome resources, and the aboveground biomass peaked in August. Then the plants started to senesce, transferring a large portion of the aboveground biomass to the belowground organs. Similar seasonal variation patterns are documented in previous studies (Asaeda, Nam, & Hietz, 2002; Windham et al., 2003). Compared with Windham's results, we found an equivalent maximal aboveground biomass of *P. australis* (1246 g DW /m<sup>2</sup> in Dongtan vs. 1117 g DW/m<sup>2</sup> in Hackensack), though under a Mediterranean climate, the peak of aboveground biomass in *P. australis* could reach 2296 g DW/m<sup>2</sup> (Asaeda, 2002). The aboveground biomass of *S. alterniflora* was obviously greater in Dongtan wetland than in the Hackensack Meadowlands (2795 g DW/m<sup>2</sup> in Dongtan vs. 679 g DW/m<sup>2</sup> in Hackensack). Therefore, aboveground primary production varies widely in salt marsh plants from different sites (Turner, 1976; Ibañez, Curco, Day, & Prat, 2002).

Our study found that the aboveground biomass of *S. alterniflora* was significantly greater than both *P. australis* and *S. maritima*. Chen et al (2003) have also found that *S. alterniflora* had a much greater primary productivity than *S. maritima* and *P. australis* in the Yangtze River estuary. In less

than 10 years, exotic *S. alterniflora* has invaded the salt marsh of the Yangtze River estuary and is rapidly replacing the native plants, *P. australis* and *S. mariqueter*. Chen, Ma and Li (2003) have shown that *S. alterniflora* occupied 13.2% of the Jiuduansha Island's area in autumn 2002, while its monocultures contributed 25.3% to the total plant biomass production of the islands. Thus, the *S. alterniflora* invasion would significantly increase the primary productivity of the salt marsh ecosystem in Yangtze River estuary and potentially have further influence on the rest of the ecosystem.

Currently, we don't know the mechanism that allows *S. alterniflora* to invade the salt marsh and replace the native plants in the Yangtze River estuary. Recent studies demonstrate that available N can affect the patterns of plant community diversity and succession in a salt marsh (Levine, Hacker, Harley, & Bertness, 1998; Rozema et al., 2002). As a result of the physiological differences in growth rate, plant growth form and plant morphology between plant species, plants that assimilate N well will dominate salt marshes (Rozema et al., 2002). Therefore, we hypothesize that there may be two possible mechanisms that allow *S. alterniflora* to invade the salt marsh and replace native plants in the Yangtze River estuary: Firstly, *S. alterniflora* can more rapidly and efficiently assimilate inorganic N than the two native species; secondly, the exotic can improve the mineralization rate and bioavailability of N in the sediment of salt marsh. This hypothesis needs to be tested further.

Bioavailabilities of nutrients and metals are affected by a wide range of environmental characteristics of the sediments, most notably the pH and redox potential (Sundareshwar, Morris, Koepfler, & Fornwalt, 2003; Cacador et al., 2000; Burke, Weis, & Weis, 2000). In coastal salt marshes, the sediments tend to be reduced. The anoxic condition may restrain N mineralization and facilitate denitrification. Thus, N is conventionally thought to limit primary production in coastal salt marshes. (Sundareshwar et al, 2003; Teal & Howes, 2002). Phosphate can be present in the form of iron, aluminium and calcium oxides or adsorbed on the surface of minerals and organic materials. Among these forms, calcium-bound P is the most important fraction in the salt marsh of the Yangtze River estuary (Gao, Xu, & Zhang, 2001). Sundareshwar et al. (2003) found that P possibly limited the N-transforming bacteria communities and would affect carbon fixation, storage and primary production in some circumstances. In a reduced condition, most of the metal often binds to sulfides in the sediment. This form of metals is unavailable to plant uptake (Weis & Weis, 2004; Williams, 2002). Salt marsh macrophytes can oxidize the sediment in the root zone through the movement of oxygen downwards through aerenchyma tissue, however, which changes the sediment Eh and pH condition.

At the same time, this oxidation can increase the magnitude of nutrient cycling and improve the availability of nutrients and metals in wetland sediment (Weis & Weis, 2004; Moorhead & Reddy, 1988). Additionally, salt marsh macrophytes may act as a conduit for the movement of nutrients and toxic metals from the sediment to the surface and near coastal seas (Burke et al., 2000).

The roots of salt marsh plants can accumulate high concentrations of metals, but only a low proportion of the metal is allocated to aerial tissues, which indicates a restriction of transport upwards (Weis & Weis, 2004; Windham et al., 2003). Similar to previous studies, our results demonstrate that metal concentrations in the aboveground tissues are lower than in belowground tissues. It is accepted that salt marsh plants possibly employ three strategies to translocate metals to aboveground tissues: the excluder strategy, indicator strategy and accumulator strategy (Baker, 1981; Windham et al., 2003). Many studies show that *P. australis* and *S. alterniflora* function as excluders. The shoot concentrations of metal are kept low until a critical soil concentration is reached, toxicity ensues, and unrestricted metal transport results (Alberts, Price, & Kania, 1990; Peverly, Surface, & Wang, 1995).

Our study shows a relatively consistent seasonal variation pattern of N and P concentrations in the aboveground tissues of three salt marsh plants. The concentrations of TN and TP in the aboveground tissues decreased gradually across the growing season. Roman and Daiber (1984) presented similar results: the relative N concentration of standing live *S. alterniflora* tissue was at a peak during the spring, and there was a steady decline until a minimum was reached at the end of the growing season. The variation patterns of N and P in the aboveground tissues were correlated with the growth dynamic of the salt marsh plants. In the early stage of the growing season, a high nutrient proportion was found to accumulate in the newly emerged photosynthetic tissue (stem and leaves) of the macrophytes. The concentrations of N and P in the aboveground tissues decreased gradually with the growth of salt marsh plants. The nutrients in the aboveground tissue were slowly translocated to the belowground tissue as the plants started to senesce (Roman & Daiber, 1984; White, 1994).

In regards to the seasonal pattern of metal concentrations, our results are consistent with patterns observed elsewhere (Gleason, Drifmeyer, & Zieman, 1979; Heller & Weber, 1998; Cacador et al., 2000). Two studies (Gleason et al., 1979; Heller & Weber, 1998) discuss decreasing concentrations of metals in the leaf tissue of *S. alterniflora* across the growing season. The decline is attributed to a growth dilution effect. Additionally, Cacador et al. (2000) suggest that the seasonal variations in root metal concentrations of *Spartina maritima* and *Halimione portulacoides* are not linearly correlated to

the biomass increment and that the greatest increase in metals occurs at the beginning of the growth period (March). This may be the result of modifying the chemistry of the rhizosphere during the plants' growth periods and consequently changing the metal availability at the interface of plant root-sediment. The same seasonal variation patterns in metal levels were also observed in other salt marsh plants, such as *Aster tripolum*, *Puccinellia maritime* and *Spartina anglica* (Leendertse, Scholten, & van Wal, 1996). But several other studies show different seasonal variation in metal concentrations in salt marsh plants. For example, Drifmeyer and Redd (1981) note that standing dead *S. alterniflora* leaves had higher metal concentrations of Mn, Fe, Cu, Zn and Ni than the leaves of live plants, whereas *P. australis* showed a different seasonal pattern with metals (Larsen & Schierup, 1981). At this point, it is difficult to generalize about seasonal changes in metal levels (Weis & Weis, 2004).

*P. australis* concentrates a greater proportion of its body-burden of metals in the root and rhizome tissue than *S. alterniflora* (Burke et al., 2000; Weis & Weis, 2004; Windham, Weis, & Weis, 2001). Kraus (1988) reported that *P. australis* had higher root concentrations of Ni, Cadmium and Pb than *S. alterniflora*, and leaf levels of the metals were comparable in the two plants. In general, our study demonstrates that the metal concentrations of plant tissues (aboveground and belowground) were greater in *S. alterniflora* than in *P. australis*, which may be related to the difference in root system and metal accumulation pattern of the two plant species (Breteler & Teal, 1981; Weis & Weis, 2004; Ye et al., 1997). The larger root system and increased number of fine roots of *S. alterniflora* compared with *P. australis* can oxidize the rhizosphere to a greater extent and increase the availability of metals. Breteler and Teal (1981) found that the mercury concentration in *S. alterniflora* increased when growing in mercury-contaminated sediment, while different populations of *P. australis* from a contaminated site and the reference marsh had a similar metal uptake of Zn, Pb and cadmium (Ye et al., 1997). Windham et al. (2001) also show a fundamental difference in Pb allocation and accumulation pattern of *S. alterniflora* and *P. australis*. *S. alterniflora* allocated significantly more of the pool to aboveground biomass than *P. australis*.

In this study, the aboveground N pools in August were 19.9 g N /m<sup>2</sup> for *P. australis*, 24.4 g N /m<sup>2</sup> for *S. alterniflora*, and 8.4 g N /m<sup>2</sup> for *S. maritima*, which were equivalent to a N assimilation rate of 199 kg N/(ha·yr), 244 kg N/(ha·yr), and 84 kg N/(ha·yr). Our results are similar to those observed in other studies. In the salt marsh of the Wadden sea area, the rate of N assimilation is 95~230kg N/(ha·yr), average 160 kg N/(ha·yr) (Rozema et al., 2002). In an Australian treatment pond, the peak

aboveground pools were 18.1 g N /m<sup>2</sup> and 1.4 g P /m<sup>2</sup> (Adcock & Ganf, 1994). In the salt marsh of the Tingitan peninsula, Morongo, *P. australis* retained N at a rate of 25.3g/m<sup>2</sup> and P at 2.76 g/m<sup>2</sup> in the aboveground tissues (Ennabili, Ater, & Radoux, 1998).

The aboveground size of nutrient pools were mainly attributed to the biomass of the salt marsh plants. Maximum nutrient accumulations in the aboveground shoots corresponded to peak biomass, which occurred in mid to late summer. Similar trends were generally observed in two Delaware Bay tidal marshes (Roman & Daiber, 1984). As a result of the significant difference in the size of nutrient pools between the exotic and native plants, the exotic plant invasion may significantly influence the biogeochemistry process of the salt marsh ecosystem. Windham (1999) examined the extent to which a change in plant species altered N cycling in a salt marsh. His results suggest that *P. australis* invasions appear to increase the magnitude of N cycles within salt marshes by sequestering more N through plant and litter uptake, stimulating the soil mineralization rates, increasing the rates of coupled nitrification-denitrification in the sediment, and generating a shift of the dominance of N export pathways—from the export of available N (ammonium) to the export of unavailable N (N<sub>2</sub>) to the atmosphere. Our results show that the aboveground nutrient pools of *S. alterniflora* were significantly greater than *P. australis* and *S. maritima* throughout the growing season as result of greater aboveground biomass, which shows that the *S. alterniflora* invasion significantly improved the rate of N and P assimilation and possibly increased the magnitude of nutrient cycles and altered the nutrient flux between the salt marsh and estuary.

Although most of the metals taken into the salt marsh plants remain belowground, amounts sufficient for concern are transported to the aboveground tissues (Windham et al., 2003; Burke et al., 2000). By comparing with the aboveground metal pools of three salt marsh plants, we found that the aboveground metal pools of *S. alterniflora* were significantly greater than the two native species. This result is mainly the result of greater net primary production in plots of *S. alterniflora*. Many studies compare the aboveground pool sizes of trace metals in *P. australis* and *S. alterniflora* under field conditions (Weis, & Weis, 2004; Windham, Weis, & Weis, 2003) and have found that *P. australis* appears to have a tighter restriction on the upward movement of metals, while *S. alterniflora* transports greater amounts to the aboveground tissues, especially the leaves. Windham et al. (2001) demonstrate that the replacement of *S. alterniflora* with *P. australis* may reduce Pb bioavailability by sequestering a greater proportion of its metal burden in the belowground tissues, which are likely to be

permanently buried. Later, they found that the document replacement of *S. alterniflora* with invading populations of *P. australis* would lead to a decrease in mercury and chromium export from the aboveground biomass into the water column and the detrital pathway in low salt marshes and a possible increase in Cu and Zn export (Windham et al., 2003). Our study demonstrates that a *S. alterniflora* invasion will possibly increase the import of Cu, Zn and Pb from the sediment into the estuarine water column and detrital food web in the Yangtze River estuary.

*S. alterniflora* can release large amount of metals through the excretion of metal-containing salts produced by leaf salt glands (Burke et al., 2000; Weis et al., 2002). Kraus (1988) estimated that *S. alterniflora* had the theoretical potential to export 145 g cadmium, 260 g Pb, 104 g chromium, 260 g Cu and 988 g nickel per ha/year through salt excretion. Greater metal release at higher salinities was observed as a result of more salt excretion. Therefore, it is certain that *S. alterniflora* can release larger quantities of metals into the marsh environment than native plants. When salt marshes flood, metals excreted from the leaves are resolubilized and can dissolve in the water column, which may influence the metal flux between a salt marsh and estuary (Marinucci, 1982; Weis et al., 2002).

In addition, Drifmeyer, Cross and Zieman (1982) report that metals from *S. alterniflora* detritus are extracted quickly and may be in a form readily available to consumers. Furthermore, two studies (Heyes, Moore, & Rudd, 1998; Helller & Webber, 1998), show a more toxic methylated form of mercury was present in *S. alterniflora*, and the percent of methylmercury in the total mercury increased from May to July (up to 48%). They speculate that either *S. alterniflora* was capable of methylating mercury, or there was a selective uptake from sediments. Relatively clean *Spartina* detritus may act primarily as a cation exchanger, adsorbing metals from sediments (Giblin, Bourg, Valiela, & Teal, 1980).

Consequently, an invasion of *S. alterniflora* may promote metal bioavailability by transporting greater amounts of metals into the aboveground tissue—readily decomposable tissues—and releasing more metals to salt into the environment through the leaves (Burke et al., 2000; Weis et al., 2002). When the aboveground tissues die and turn into detritus, metal stored in the tissues are likely to become available to deposit feeders and enter the estuarine food web (Weis, Windham, Santiago-Bass, & Weis, 2002; Windham et al., 2003). Therefore, wetland managers and ecologists should be concerned with the effects of *S. alterniflora* invasions on the biogeochemistry processes of salt marsh ecosystems and particularly take measures to control the spreading of *S. alterniflora* in the Yangtze

River estuary.

This study suggests that invasive *S. alterniflora* holds greater aboveground element pools than the two native plants and infers that its invasion will significantly increase the magnitude of nutrient cycling and bioavailability of metals in the salt marsh and transport more metals into the water column and the detrital food web in the estuary, which is harmful for the health of the ecosystem. To prove this position, three areas require further study: (1) the dynamic change of nutrients and metal concentrations in the litter decomposition process of three salt marsh plants, (2) whether consumption of contaminated detritus has effects on macro invertebrates and nekton, and (3) the influence of *S. alterniflora* invasion on the bioavailabilities of nutrients and metals in the sediment and element flux between the salt marsh and estuary.

## 5. Conclusion

During the growing season of 2005, we studied the seasonal patterns of the aboveground biomass accumulation, tissue element concentration, and aboveground element pools of three dominant salt marsh macrophytes in the Yangtze River estuary. The aboveground biomass was significantly greater in plots of *S. alterniflora* than in plots of *P. australis* and *S. mariqueter* throughout the season, which shows that the *S. alterniflora* invasion significantly increased the aboveground primary productivity of the salt marsh ecosystem. The variation patterns of the nutrients and metals in the aboveground tissues were similar and showed a decreasing trend from April to October. Furthermore, the aboveground pools of nutrients and metals were consistently greater in the plots of *S. alterniflora* than *P. australis* and *S. mariqueter*. We suggest that the rapid replacement of the native plants (*P. australis* and *S. mariqueter*) by the exotic plant (*S. alterniflora*) will significantly increase the magnitude of nutrient cycling and bioavailability of metals in the salt marsh, transport more trace metals into the water column and the detrital food web, and further influence the biogeochemistry process of the Yangtze River estuary. Further study of the dynamic change of nutrient and trace metal concentrations in the process of salt marsh plant litter decomposition and the influence of exotic plant invasions on the biogeochemistry cycling process of salt marsh ecosystems is necessary.

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Table 1 Linear regressions of TN and TP against time calculated for three salt marsh macrophytes. Covariance analysis tests the difference among the slopes of three species, and  $r^2$  estimates the proportion of the total variation explained by the regression model.

Factors	Plants	Regression analysis			Covariance analysis	
		Intercept	Slope	$r^2$	$F$ ratio	$P$
Aboveground TN	<i>Phragmites australis</i>	0.0231	-0.0015	0.58**	0.354	NS
	<i>Spartina alterniflora</i>	0.0229	-0.0017	0.86**		
	<i>Scirpus mariqueter</i>	0.0239	-0.0011	0.07		
Aboveground TP	<i>Phragmites australis</i>	0.0035	-0.0003	0.79**	2.714	NS
	<i>Spartina alterniflora</i>	0.0045	-0.0004	0.73**		
	<i>Scirpus mariqueter</i>	0.003	-0.0002	0.21		
Belowground TN	<i>Phragmites australis</i>	0.0098	-0.0003	0.21	7.058	**
	<i>Spartina alterniflora</i>	0.0078	-0.00034	0.15		
	<i>Scirpus mariqueter</i>	0.0219	-0.0019	0.17		
Belowground TP	<i>Phragmites australis</i>	0.0012	$3.19 \times 10^{-5}$	0.05	1.015	NS
	<i>Spartina alterniflora</i>	0.0015	$-8.60 \times 10^{-5}$	0.24		
	<i>Scirpus mariqueter</i>	0.0033	-0.00018	0.15		

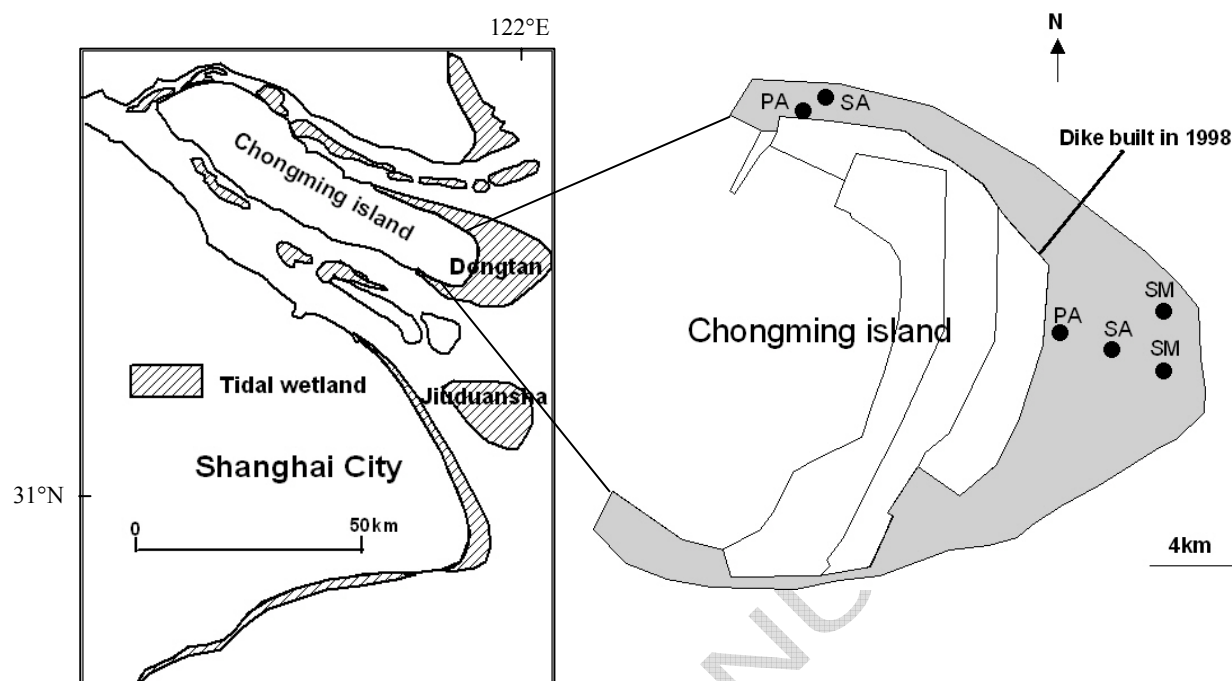
\*\*= $P < 0.01$ ; \*= $P < 0.05$ ; NS=  $P > 0.05$

Table 2 Linear regressions of Cu, Zn and Pb concentrations against time calculated for three salt marsh macrophytes. Covariance analysis tested the difference among the slopes of three plant species, and  $r^2$  estimates the proportion of the total variation explained by the regression model.

Factors	Plants	Regression analysis			Covariance analysis	
		Intercept	Slope	$r^2$	F ratio	P
Aboveground Cu	<i>Phragmites australis</i>	0.0231	-0.0015	0.58**	2.75	NS
	<i>Spartina alterniflora</i>	35.9800	-3.6317	0.79**		
	<i>Scirpus mariqueter</i>	12.9297	-0.0763	0.21		
Aboveground Zn	<i>Phragmites australis</i>	157.1804	-16.12	0.05	5.031	*
	<i>Spartina alterniflora</i>	110.7700	-8.9935	0.86**		
	<i>Scirpus mariqueter</i>	83.8454	-5.7074	0.73**		
Aboveground Pb	<i>Phragmites australis</i>	18.3300	-1.7289	0.15	3.575	*
	<i>Spartina alterniflora</i>	15.3895	-1.0884	0.24		
	<i>Scirpus mariqueter</i>	2.9675	0.3382	0.07		
Belowground Cu	<i>Phragmites australis</i>	84.6622	-8.1062	0.21*	2.719	NS
	<i>Spartina alterniflora</i>	125.2146	-11.6098	0.67**		
	<i>Scirpus mariqueter</i>	38.5035	-2.5135	0.15		
Belowground Zn	<i>Phragmites australis</i>	173.3475	-15.58	0.58**	1.901	NS
	<i>Spartina alterniflora</i>	291.9246	-24.3172	0.79**		
	<i>Scirpus mariqueter</i>	174.9108	-13.2795	0.21		
Belowground Pb	<i>Phragmites australis</i>	39.4440	-3.7542	0.05	6.755	**
	<i>Spartina alterniflora</i>	29.8884	-2.5782	0.86**		
	<i>Scirpus mariqueter</i>	-1.8796	1.2606	0.14		

\*\*= $P < 0.01$ ; \*= $P < 0.05$ ; NS=  $P > 0.05$

582 Figure1



583  
 584 Fig.1 Yangtze River estuary and salt marsh site studied. The black circles show the sampling sites.  
 585 The abbreviation: PA-the sampling sites in the monocultures of *Phragmites australis*, SA-the  
 586 sampling sites in the monocultures of *Spartina alterniflora*, SM-the sampling sites in the monocultures  
 587 of *Scirpus mariqueter*. In each sampling site, three 1m<sup>2</sup> plots were deployed for collecting plant  
 588 sample.

Figure 2

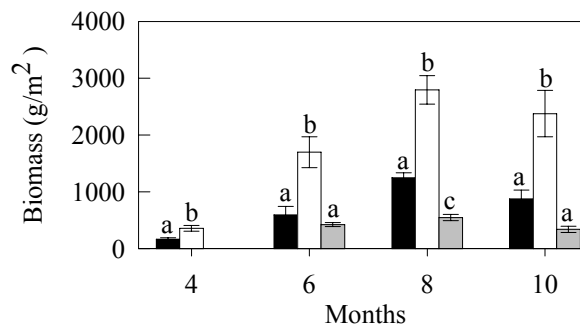


Fig.2 Aboveground biomasses of three salt marsh macrophytes throughout growing season (April-October). The bars denote  $\pm 1$  standard error ( $n=6$ ). ■ *Phragmites australis*, □ *Spartina alterniflora*, ■ *Scirpus mariqueter*. Different lowercase letters denote significant differences between plant species at  $\alpha < 0.05$ .



Figure 3

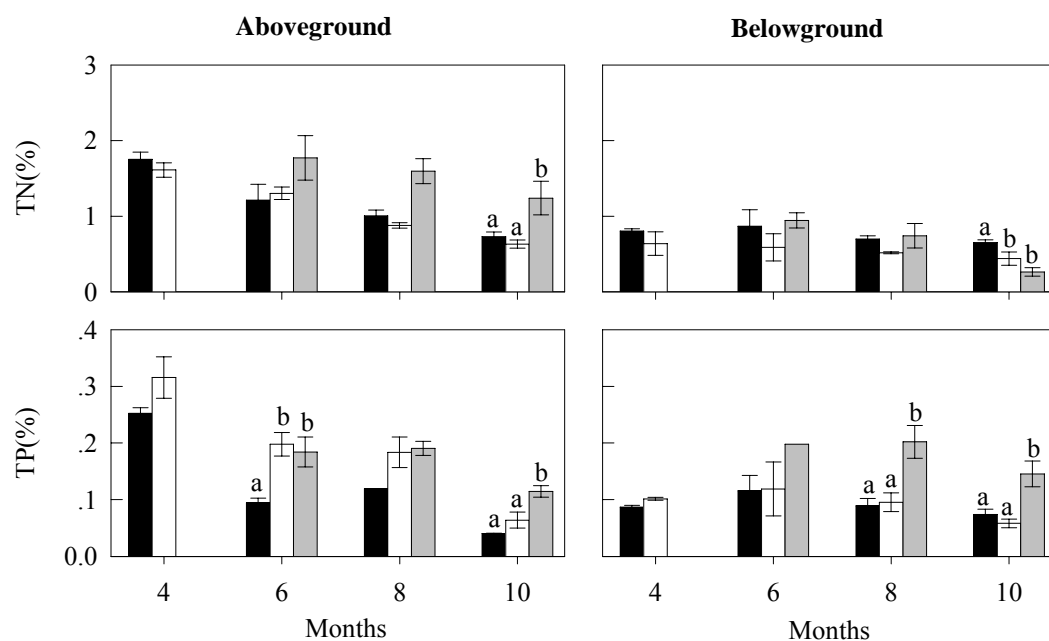


Fig.3 Concentrations of TN and TP in aboveground and belowground tissues of three salt marsh macrophytes throughout growing season (April-October). The bars denote  $\pm 1$  standard error (n=6). ■ *Phragmites australis*, □ *Spartina alterniflora*, ▒ *Scirpus mariqueter*. Different lowercase letters denote significant differences between plant species at  $\alpha < 0.05$ .

Figure 4

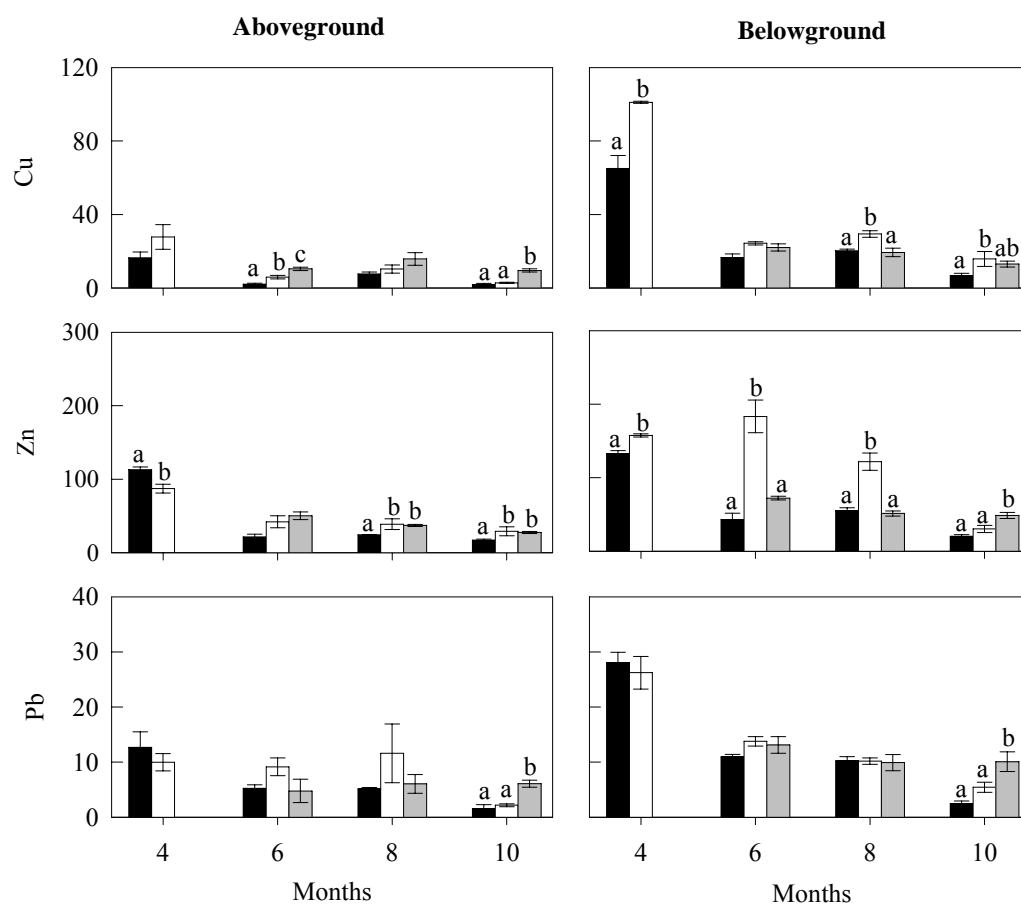
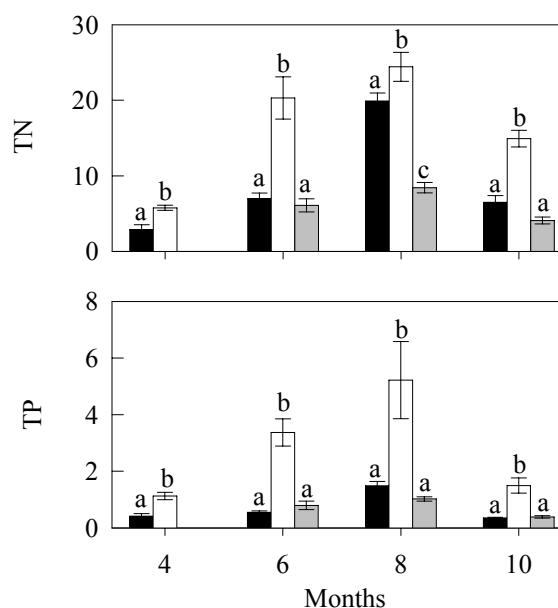


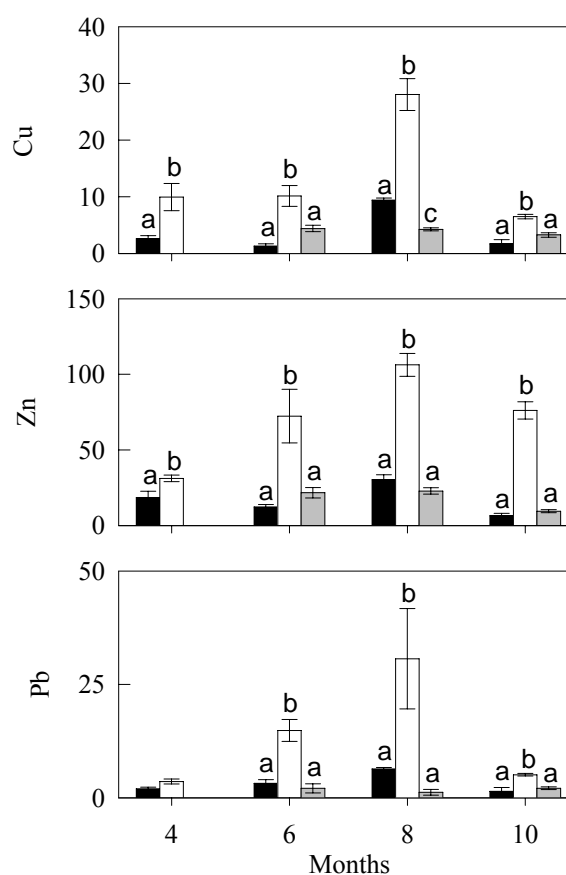
Fig.4 Concentrations of heavy metals (mg/kg dry weight) in aboveground and belowground tissues of three salt marsh macrophytes throughout growing season (April-October). The bars denote  $\pm 1$  standard error (n=6). ■ *Phragmites australis*, □ *Spartina alterniflora*, ■ *Scirpus marigueter*. Different lowercase letters denote significant differences between plant species at  $\alpha < 0.05$ .

667 Figure 5



668  
 669 Fig.5 Aboveground TN and TP pools (g/m<sup>2</sup>) of three salt marsh macrophytes throughout growing  
 670 season (April-October). The bars denote  $\pm 1$  standard error (n=6). ■ *Phragmites australis*, □ *Spartina*  
 671 *alterniflora*, ■ *Scirpus mariqueter*. Different lowercase letters denote significant differences between  
 672 plant species at  $\alpha < 0.05$ .

686 Figure 6



687  
 688 Fig.6 Aboveground metal pools (mg/m<sup>2</sup>) of three salt marsh macrophytes throughout growing season  
 689 (April-October). The bars denote  $\pm 1$  standard error (n=6). ■ *Phragmites australis*, □ *Spartina*  
 690 *alterniflora*, ■ *Scirpus maritimus*. Different lowercase letters denote significant differences between  
 691 plant species at  $\alpha < 0.05$ .