

# Growth and root distribution of *Vallisneria natans* in heterogeneous sediment environments

Yonghong Xie<sup>a,\*</sup>, Wei Deng<sup>b</sup>, Jinda Wang<sup>c</sup>

<sup>a</sup> Institute of Subtropical Agriculture, The Chinese Academy of Sciences, Changsha 410125, PR China

<sup>b</sup> Institute of Mountain Hazards and Environment, The Chinese Academy of Sciences, Chengdu 610041, PR China

<sup>c</sup> Northeast Institute of Geography and Agricultural Ecology, The Chinese Academy of Sciences, Changchun 130012, PR China

Received 31 August 2005; received in revised form 9 June 2006; accepted 10 August 2006

## Abstract

Plant growth, biomass allocation and root distribution were investigated in the submerged macrophyte *Vallisneria natans* growing in heterogeneous sediments. Experimentally heterogeneous sediment environments were constructed by randomly placing 4 cm of clay or sandy loam into the top (0–4 cm) or bottom (4–8 cm) layer within an experimental tray, providing two homogeneous and two heterogeneous treatments. Biomass accumulation was significantly affected by the experimental treatments: higher in the homogeneous sediment of clay (32 mg per plant) and the two heterogeneous treatments (about 27 mg per plant), but lower in the homogeneous sediment of sandy loam (15 mg per plant). Root: shoot ratio was also different among the four treatments. Compared with the treatments of clay in the top layer, plants allocated more biomass to roots at the treatments of sandy loam in the top layer. Heterogeneous sediments significantly affected root distribution pattern. Compared with the treatments of sandy loam in the bottom layer, root number (7–8 versus 13–14) and total root length (3.6–4.0 cm versus 29.5–40.0 cm) in the bottom layer were significantly higher in the treatments with clay in the bottom layer. These results indicate that both sediment structure and nutrient availability influence growth and root system distribution of *V. natans*.

© 2006 Elsevier B.V. All rights reserved.

**Keywords:** Heterogeneous environment; Nutrient; phenotype; Root foraging; *Vallisneria natans*

## 1. Introduction

Nutrient availability in natural habitats is heterogeneous both in space and time, even on a small scale (Jackson and Caldwell, 1993; Gross et al., 1995; Xie et al., 2004). In terrestrial systems, nutrient patches are often found, and plant responses can include changes in biomass, in clonal characteristics, in uptake kinetics, in root morphology and in root architecture (Gross et al., 1995; Einsmann et al., 1999; Šmilauerová and Šmilauer, 2002). Studies have shown that clonal foraging, by placing ramets into the nutrient patches (Hutchings and de Kroon, 1994; Dong and Alaten, 1999), and root foraging, by proliferating plant roots in nutrient-enriched patches relative to nutrient-poor ones (Linkohr et al., 2002; Zhang and Forde, 1998), can substantially enhance nutrient acquisition. Root system responses to the spatial and temporal distributions of nutrients in infertile environments are predicted

to be critical determinants of species survival (Jackson and Caldwell, 1989).

In freshwater systems, nutrient heterogeneity may be different from terrestrial systems. One difference is that nutrient layering might be more pronounced in freshwater systems (Klump and Martens, 1981, 1989), because of water-level fluctuation, flooding and variation in the sediment load in the water. In contrast to terrestrial systems, aquatic systems may have nutrient-poor sediments lying on the top. As a result, stratification in sediments can often be observed in natural aquatic habitats. Therefore, the response of submerged macrophytes to heterogeneous nutrient environments might also differ from terrestrial plants. Submerged macrophytes respond to sediment nutrient availability by modifying root morphology and root: shoot ratio (Mantai and Newton, 1982; Barko et al., 1991; Xie et al., 2005), but nothing is known about the response to sediment heterogeneity.

*Vallisneria natans* (Lour.) Hara, a perennial submerged clonal plant with a wide geographical range, can be found in different freshwater habitats in China. This plant has a highly adaptive capability, and can grow in different types of sediment. The root

\* Corresponding author. Tel.: +86 731 4615203; fax: +86 731 4615203.

E-mail address: [yonghongxie@163.com](mailto:yonghongxie@163.com) (Y. Xie).

system of this plant is made up of adventitious roots without lateral roots (Xie et al., 2005). We have certified before that root morphology of this plant is significantly affected by sediment type (Xie et al., 2005), and the aim of the present experiment is to elucidate the effects of sediment structure on plant growth and root distribution. To this end, young seedlings of *V. natans*, germinating from seeds, were grown in constructed heterogeneous sediments to answer the following questions. Firstly, does sediment distribution pattern affect plant growth? Secondly, does this plant adjust its biomass allocation and root distribution pattern as a response to sediment heterogeneity?

## 2. Materials and methods

### 2.1. Plant materials and plant culture

Seeds of *V. natans* were collected from Liangzi Lake (30°6′–30°18′N, 114.24′–114°36′E), a nutrient-poor lake in Hubei Province, China, at the end of December, 2003, after which they were placed in an indoor plastic bin (50 cm × 40 cm × 30 cm) with 3 cm of water. Some seeds were transported to Nanjing University in early March 2004, then were placed into six plastic trays (22 cm × 12 cm × 8 cm) with 5 cm of water and 3 cm of horticultural soil for culture of young seedlings. Several days later, the seeds began to germinate, and we placed the trays into a greenhouse, where the temperature was controlled at  $25 \pm 2$  °C in the day and  $15 \pm 2$  °C at night and light was provided by metal halide bulbs at a photo flux density of  $300 \mu\text{mol m}^{-2} \text{s}^{-1}$  (PAR) in a 14 h light/10 h dark cycle. Due to high density and limited water volume in plastic trays, well water was replaced completely on a daily basis to prevent the negative impacts of phytoplankton and algae and to assure normal growth of the young seedlings. One month later, the trays were transported into a plastic bin (70 cm × 50 cm × 40 cm), and well water was replaced every 2 days.

Thirty-two young seedlings (as genets) with similar size (three leaves, about 3–4 cm in height) were transplanted into 32 plastic trays (22 cm × 12 cm × 8 cm, one plant per tray). The trays were then placed into four plastic bins (70 cm × 50 cm × 40 cm, eight trays per bin) to start the experiment. The experiment was performed in the same greenhouse as seedling culture. Tap water was supplied timely according to the growth of the plants during the experimental period. The final depth was 40 cm.

### 2.2. Experimental set-up

We divided the experimental trays into two equal layers: the top (0–4 cm depth) and bottom layer (4–8 cm depth) by a nylon tent netting with 1 mm mesh, and each layer was loaded with 4 cm of sediment (Fig. 1). Root diameter of *V. natans* is about 0.3–0.4 mm (Xie et al., 2005), so a netting with 1 mm mesh can assure free rooting. Two types of sediment, clay and sandy loam, were used in this experiment, providing a total of four treatments: homogeneous sediment of clay (CC), clay in the top layer and sandy loam in the bottom layer (CS), sandy loam in the top layer and clay in the bottom layer (SC), homogeneous sediment of sandy loam (SS). Each plastic bin contained all treatments (two trays per treatment), so each treatment was replicated eight times. Treatments were allocated randomly over the bins, and bins thus served as blocks. The sandy loam was collected from a local river (Qinhuai River), containing  $8.3 \mu\text{g g}^{-1}$  exchangeable N and  $2.5 \mu\text{g g}^{-1}$  exchangeable P. The clay was collected from Taihu Lake (30°5′–32°8′N, 119°8′–121°6′E), a highly eutrophic lake in Jiangsu Province, China, containing  $103 \mu\text{g g}^{-1}$  exchangeable N and  $36 \mu\text{g g}^{-1}$  exchangeable P.

### 2.3. Root distribution

The plants were harvested after they were grown for 21 days, when the largest plant was to generate new ramets. We excavated the sediments by hand carefully, then gently washed the sediment in the bottom layer by tap water, after which root number was counted and each root length was measured by a vernier caliper. The plants were washed carefully with tap water, then were divided into leaves, roots, stolons and rhizomes, and weighted as fresh weight. Specific root length was calculated as root length divided by root biomass. For each measurement, 5–7 roots were randomly chosen from different plants. We replicated six times for each treatment. Finally, plant parts were oven dried at 85 °C for 24 h, and dry weight recorded. All fresh weights were then converted into dry weight. Total root length was estimated by multiplying total root weight by specific root length. Root length in the bottom layer was defined as the sum of all root length in 4–8 cm depth, and root length in the top layer was calculated by subtracting root length in the bottom layer from total root length. Root length distribution ratio in the top or bottom layer was defined as the ratio of root length in the top or bottom layer to total root length (expressed as %).

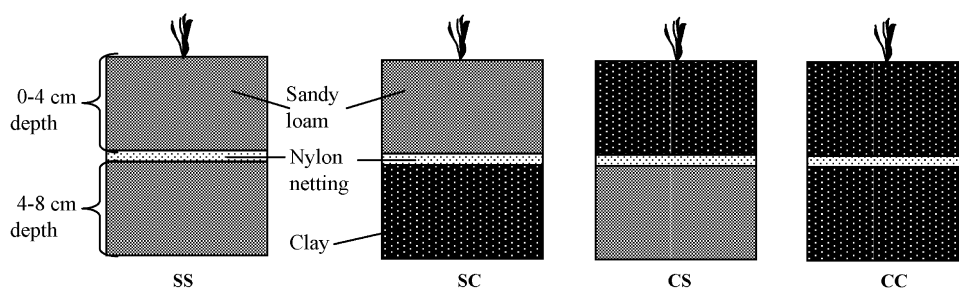


Fig. 1. Configurations of plastic trays to test root distribution of the submerged macrophyte *Vallisneria natans* grown in heterogeneous sediments for 21 days. SS indicates sandy loam in 0–4 and 4–8 cm depth; SC indicates sandy loam in 0–4 cm depth and clay in 4–8 cm depth; CS indicates clay in 0–4 cm depth and sandy loam in 4–8 cm depth; CC indicates clay in 0–4 and 4–8 cm depth.

## 2.4. Statistical analyses

Treatment effects on biomass accumulation, biomass allocation, root number and length in the bottom layer were analyzed by one-way ANOVA using the software SPSS 12.0 for Windows. Multiple comparisons of means were performed by Duncan's test at the 0.05 significance level. Data were  $\log_{10}$ -transformed if necessary to reduce heterogeneity of variances, and homogeneity was tested using Levene's test. In order to separate treatment effects from plant size effects on root distribution, one-way ANCOVA was used, with treatment as main factor, biomass as covariate and root number and length in the bottom layer as dependent factors. Data were  $\log_{10}$ -transformed.

## 3. Results

### 3.1. Biomass accumulation

Biomass accumulation was significantly affected by the experimental treatments (Fig. 2). Total biomass at harvest time

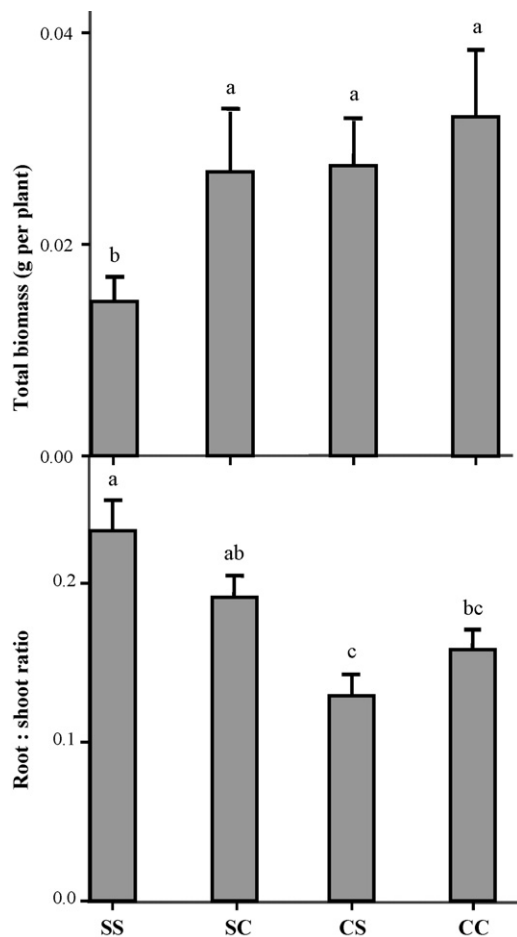


Fig. 2. Biomass accumulation and root: shoot ratio (means  $\pm$  S.E.,  $n = 8$ ) of *Vallisneria natans* grown in four treatments for 21 days. SS indicates sandy loam in 0–4 and 4–8 cm depth; SC indicates sandy loam in 0–4 cm depth and clay in 4–8 cm depth; CS indicates clay in 0–4 cm depth and sandy loam in 4–8 cm depth; CC indicates clay in 0–4 and 4–8 cm depth. Different letters indicate significant difference among treatments ( $P < 0.05$ ).

was higher in the CC (32 mg per plant) and the two heterogeneous sediment treatments (about 27 mg per plant), and lower in the SS treatment (15 mg per plant). However, there were no significant differences among CC, SC and CS treatments.

### 3.2. Biomass allocation

Biomass allocation was significantly affected by the experimental treatment (Fig. 2). Of the four treatments, root: shoot ratio was highest in the SS treatment (0.18), intermediate in the SC and CC treatments (0.13–0.14), and lowest in the CS treatment (0.11). Sandy loam in the top layer usually resulted in a high root: shoot ratio.

### 3.3. Root distribution

Root number and length in the bottom layer were significantly affected by experimental treatments (Fig. 3). Root number was 14 in the SC treatment, 13 in the CC treatment and about seven in the SS and CS treatments. Root length showed the same tendency

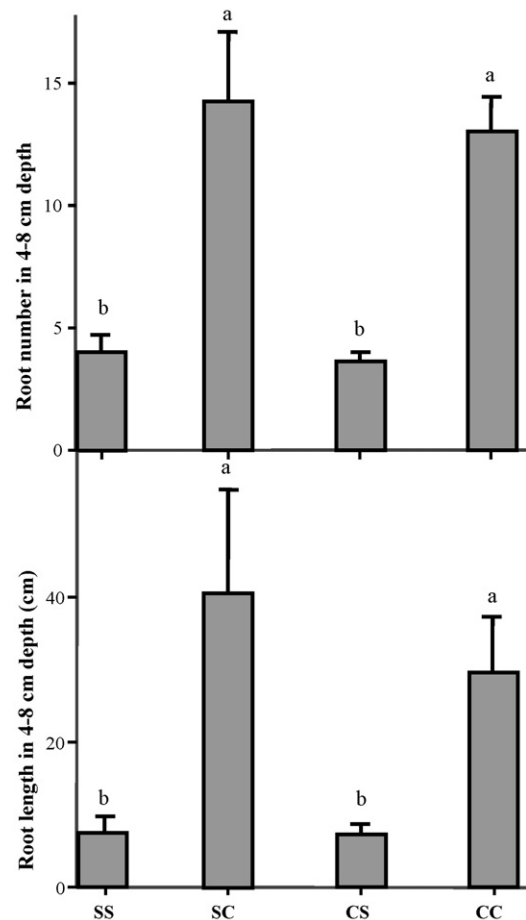


Fig. 3. Root number and root length (means  $\pm$  S.E.,  $n = 8$ ) of *Vallisneria natans* in the bottom layer. SS indicates sandy loam in 0–4 and 4–8 cm depth; SC indicates sandy loam in 0–4 cm depth and clay in 4–8 cm depth; CS indicates clay in 0–4 cm depth and sandy loam in 4–8 cm depth; CC indicates clay in 0–4 and 4–8 cm depth. Different letters indicate significant differences among treatments ( $P < 0.05$ ).

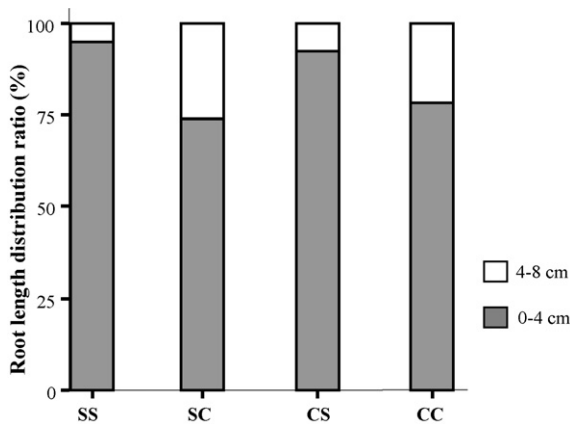


Fig. 4. Relative root length distribution ratio of *Vallisneria natans* in 0–4 and 4–8 cm of sediment depth. SS indicates sandy loam in 0–4 and 4–8 cm depth; SC indicates sandy loam in 0–4 cm depth and clay in 4–8 cm depth; CS indicates clay in 0–4 cm depth and sandy loam in 4–8 cm depth; CC indicates clay in 0–4 and 4–8 cm depth.

as root number, and was highest in the SC treatment (40 cm), intermediate in the CC treatment (30 cm), and lowest in the SS and CS treatments (3.6–4.0 cm). However, there was no significant difference in root number and length between SC and CC treatments, and between SS and CS treatments ( $P > 0.05$ , Fig. 3). Relative root length distribution percentage in the SS and CS treatments was 74–78% in the top layer and 21–26% in the bottom layer, but 92–95% in top layer and 5–8% in the bottom layer in the SC and CC treatments (Fig. 4). A one-way ANCOVA showed that both plant size and treatment had significant effects on root number and root length in the bottom layer ( $P < 0.001$ ), indicating that root system distribution was influenced by sediment structure. It is clear that more roots were

distributed in the bottom layer when the sediment was clay, but less when the sediment was sandy loam (Fig. 5).

#### 4. Discussion

More roots were distributed in the top layer of the SS and CS treatments when sediment in the bottom layer was infertile sandy loam, but more were found in the bottom layer in the CC and SC treatments when this layer was fertile clay, indicating that sediment structure and nutrient availability play an important role in regulating root system distribution of *V. natans*. In most natural freshwater sediments the aerobic sediment surface layer serves as a channel of sediment nutrient loading to water, and continual deposition onto the sediment surface of decaying organic matter (Andersen, 1982), which is an important nutrient source for the growth of submerged macrophytes by nitrification and mineralization of microbes. As a result, the sediment surface often has the highest nutrient availability (Bonser et al., 1996) when sediment in the bottom layer is infertile (such as in the SS treatment). In infertile environments, root shallowness has been viewed as a topsoil foraging behavior at minimum metabolic cost, by increasing the total absorptive surface of the root system and lowering inter-root competition (Lynch and Brown, 2001; López-Bucio et al., 2003). Therefore, concentration of root activity in the top layer can be interpreted as adaptive response to higher nutrient content in this layer. Root distribution in the CS treatment also provides direct evidence to support the results of the SS treatment. Here, less roots in the bottom layer may suggest an escape from the infertile environment. In addition, root number and length distribution in the SC treatment may suggest that *V. natans* can reach fertile sediment by penetrating 4 cm of infertile top-layer sediment. Furthermore, sediment nutrient in the bottom layer of the SC treatment can continually load to the water, and in turn increase nutrient availability in the top layer. In terrestrial plant, lateral roots often proliferate in nutrient-enriched patches relative to nutrient-poor ones (Linkohr et al., 2002; Zhang and Forde, 1998), but the response of *V. natans* was different: it concentrates root mass, but not root number. Still, our experiment clearly showed that *V. natans* distributes most of their roots in fertile sediment, which supports the concept of root foraging (Linkohr et al., 2002; Zhang and Forde, 1998).

Shifts in biomass allocation reflect an adaptive response to changed environmental conditions. Plants usually adjust biomass allocation to minimize imbalance in any critical resource in a manner that maximizes plant growth rate (Bloom et al., 1985). *V. natans* allocated relatively more biomass to roots when grown in sandy loam in the top layer, which is a typical response to an infertile environment (Bloom et al., 1985). Increase of root mass ratio substantially increases the absorptive surface area exposed to sediment when grown in sandy loam, which is favorable for nutrient capture in infertile environments (Barko et al., 1991).

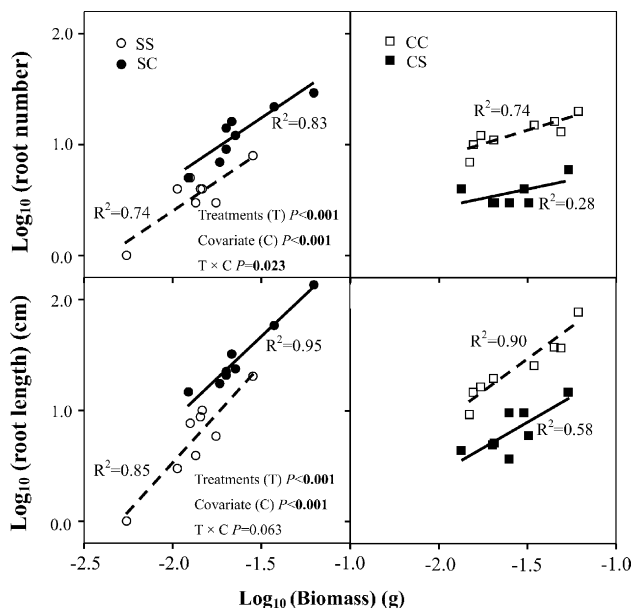


Fig. 5. Influences of heterogeneous sediments on the allometry of root number and total rootlength of *Vallisneria natans* in 4–8 cm of sediment depth. SS indicates sandy loam in 0–4 and 4–8 cm depth; SC indicates sandy loam in 0–4 cm depth and clay in 4–8 cm depth; CS indicates clay in 0–4 cm depth and sandy loam in 4–8 cm depth; CC indicates clay in 0–4 and 4–8 cm depth.

#### Acknowledgements

The authors greatly appreciate the comments given by Dr. Jan Vermaat and the two anonymous reviewers on an earlier

version. We also thank Dr. Shuqing An for providing experimental conditions, and Keyan Xiao for collecting seeds of *V. natans* from Liangzi Lake. This research was supported by the National Natural Science Foundation of China (30300032) and the National Basic Research Program of China (2006CB403301).

## References

- Andersen, J.M., 1982. Effect on nitrate concentration in lake water on phosphate release from the sediment. *Water Res.* 16, 411–419.
- Barko, J.W., Gunnison, G., Carpenter, S.R., 1991. Sediment interactions with submersed macrophyte growth and community dynamics. *Aquat. Bot.* 41, 41–65.
- Bloom, A.J., Chapin, F.S., Mooney, H.A., 1985. Resource limitation in plants—an economic analogy. *Annu. Rev. Ecol. Syst.* 16, 363–439.
- Bonser, A.M., Lynch, J., Snapp, S., 1996. Effect of phosphorus deficiency on growth angle of basal roots in *Phaseolus vulgaris*. *New Phytol.* 132, 281–288.
- Dong, M., Alaten, B., 1999. Clonal plasticity in response to rhizome severing and heterogeneous resource supply in the rhizomatous grass *Psammochloa villosa* in an Inner Mongolian dune. *China. Plant Ecol.* 141, 53–58.
- Einsmann, J.C., Jones, R.H., Pu, M., Mitchell, R.J., 1999. Nutrient foraging traits in 10 co-occurring plant species of contrasting life forms. *J. Ecol.* 87, 609–619.
- Gross, K.L., Pregitzer, K.S., Burton, A.J., 1995. Spatial variation in nitrogen availability in three successional plant communities. *J. Ecol.* 83, 357–367.
- Hutchings, M.J., de Kroon, H., 1994. Foraging in plants: the role of morphological plasticity in resource acquisition. *Adv. Ecol. Res.* 25, 159–238.
- Jackson, R.B., Caldwell, M.M., 1989. The timing and degree of root proliferation in fertile-soil microsites for three cold-desert perennials. *Oecologia* 81, 149–153.
- Jackson, R.B., Caldwell, M.M., 1993. Geostatistical patterns of soil heterogeneity around individual perennial plants. *J. Ecol.* 81, 683–692.
- Klump, J.V., Martens, C.S., 1981. Biogeochemical cycling in an organic rich coastal marine basin: 2. Nutrient sediment-water exchange processes. *Geochim. Cosmochim. Acta* 45, 101–122.
- Klump, J.V., Martens, C.S., 1989. The seasonality of nutrient regeneration in an organic-rich coastal sediment—kinetic modeling of changing pore-water nutrient and sulfate distributions. *Limnol. Oceanogr.* 34, 559–577.
- Linkohr, B.I., Williamson, L.C., Fitter, A.H., Leyse, H.M.O., 2002. Nitrogen and phosphorus availability and distribution have different effects on root system architecture of *Arabidopsis*. *Plant J.* 29, 751–760.
- López-Bucio, J., Cruz-Ramírez, A., Herrera-Estrella, L., 2003. The role of nutrient availability in regulating root architecture. *Curr. Opin. Plant Biol.* 6, 280–287.
- Lynch, J.P., Brown, K.M., 2001. Topsoil foraging— an architectural adaptation of plants to low phosphorus availability. *Plant Soil* 237, 225–237.
- Mantai, K.E., Newton, M.E., 1982. Root growth in *Myrophyllosum*: a specific plant response to nutrient availability. *Aquat. Bot.* 13, 45–55.
- Šmilauerová, M., Šmilauer, P., 2002. Morphological responses of plant roots to heterogeneity of soil resources. *New Phytol.* 154, 703–715.
- Xie, Y., Wen, M., Yu, D., Li, Y., 2004. Growth and resource allocation of water hyacinth as affected by gradually increasing nutrient concentrations. *Aquat. Bot.* 79, 257–266.
- Xie, Y., An, S., Yao, X., Xiao, K., Zhang, C., 2005. Short-time response in root morphology of *Vallisneria natans* to sediment type and water-column nutrient. *Aquat. Bot.* 81, 85–96.
- Zhang, H., Forde, B.G., 1998. An *Arabidopsis* MANS box gene that controls nutrient induced changes in root architecture. *Science* 279, 407–409.