PHOTOSYNTHETICA 44 (2): 202-207, 2006

Photosystem 2 activities of hyper-accumulator *Dicranopteris dichotoma* Bernh from a light rare earth elements mine

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Abstract

The distribution of rare earth elements (REEs) in the fern *Dicranopteris dichotoma* Bernh plants from a light rare earth elements mine (LRM) and a non-mining (NM) area in Longnan county of Jiangxi province, China were investigated by means of inductively coupled plasma-mass spectrometry, transmission electron microscopy, and energy-dispersive X-ray microanalysis. The photosynthetic characteristics of *D. dichotoma* were studied by chlorophyll (Chl) *a* fluorescence kinetics. Contents of REEs in the lamina and the root of *D. dichotoma* were higher than those in soils, and were mainly distributed in lamina. A part of them was found in the chloroplast. By comparing with *D. dichotoma* from NM area, the efficiency of photosystem 2 photochemistry and electron transport rate were significantly enhanced in lamina of the plant from LRM because most of REEs deposits were distributed along cell wall, in vacuole, and in chloroplast. High contents of REEs in lamina did not decrease the photosynthetic activities in LRM plants of *D. dichotoma*. Besides, *D. dichotoma* could change its β-carotene content to avoid the damaging effect of high REEs content.

Additional key words: β-carotene; chlorophyll fluorescence; photosynthetic electron transport; photosystem 2; thylakoid membrane.

Introduction

Rare earth elements (REEs), comprising lanthanides (Ln) and yttrium (Y), can be divided into the light rare earth elements (LREEs) group and heavy rare earth elements (HREEs) group according to atomic mass. The LREEs include lanthanum (La) to europium (Eu), whereas HREEs include gadolinium (Gd) to lutetium (Lu). The REEs at low concentrations (usually less than 0.5 mM) can improve plant photosynthetic efficiency, crop quality, and plant resistance to disease and stress (Xu and Xiao 1985, Chang 1991, Hu *et al*. 2002). In contrast, the REEs at high concentrations (usually more than 0.5 mM) inhibit growth of plants (Clijsters and van Assche 1985, Chu *et al*. 2000b, 2001). Some researchers show that REEs can modulate plant photosynthesis by K^+, Na^+, or Ca^{2+} (Poovaiah and Leopold 1976, David and Karlish 1991), ribulose-1,5-bisphosphate carboxylase/oxygenase (Chen *et al*. 2000a), and indolylacetic acid (Shen and Zhang 1994). Other authors suggest that REEs do not have direct effect on plant photosynthesis (Li *et al*. 1992, 1994, Zhou and Liu 1998). Whether REEs penetrate into the cytoplasm of animal or plant is a contentious question. The studies of calcium ion transport and its inhibitor in eukaryotic cells show that REEs can not penetrate into cytoplasm of the living cell (Peterson *et al*. 1986). Studies on the distribution of REEs in bacteria (Bayer and Bayer 1991, Merroun *et al*. 2003) and erythrocytes (Yi *et al*. 1999) indicate that REEs can not only penetrate into cytoplasm of bacteria and algae, which accumulate large amounts of REEs, but penetrate also into human cells.

Received 26 April 2005, *accepted* 24 October 2005.
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Abbreviations: Chl − chlorophyll; F_0 − minimal fluorescence level in dark-adapted leaves; F_m − maximal fluorescence level in dark-adapted leaves; F_v − maximum variable fluorescence level in dark-adapted leaves; F_0' − minimal fluorescence level in light-adapted leaves; F_m' − maximal fluorescence level in light-adapted leaves; F_v' − maximum variable fluorescence level in light-adapted leaves; F_v/F_m − maximal efficiency of PS2 photochemistry; F_v'/F_m' − efficiency of excitation energy capture by open PS2 reaction centres; LRM − light rare elements mine; NM − non-mining area; NPQ − non-photochemical quenching; q_P − photochemical quenching coefficient; PS2 − photosystem 2; Φ_{PS2} − actual PS2 efficiency.
Acknowledgements: This work was supported by the National Natural Science Foundation of China (39890390).

China is one of the most important countries in the production of REEs. This causes REEs pollution in vicinal places of some rare earth mining area and agricultural fields. Phytoremediation, the use of plants for environmental restoration, is an emerging cleanup technology (Lasat 2002). The fern *Dicranopteris dichotoma* Bernh which belongs to *Gleicheniaceae* is a hyperaccumulator of REEs and can be used in phytoremediation of REEs pollution (Ichihashi *et al*. 1992). REEs are distributed in root, stem, and lamina of this species. The REEs binding protein (Wang *et al.* 2003a), polysaccharides, nucleic acids (Wang *et al*. 1997, 1999), and chlorophyll (Hong *et al*. 1999) are distributed in the lamina of the plant. The possible roles of REEs in this plant are still not well explained. We studied the contents of REEs in different parts of *D. dichotoma*, their fine location in its lamina cells from LRE mine (LRM) and non-mining (NM) area in Longnan county of Jiangxi Province, China, and their effects on photosynthetic characteristics.

Materials and methods

Plants: Longnan County is located at 114°56′–114°58′E, 24°41′–24°52′N. The climate of Longnan County is warm and moist. The annual mean temperature is 18.5–19.0 °C, annual mean frost-free period 272–287 d, annual rainfall 1 439.8–1 515.6 mm, annual mean relative humidity 76–79 %, annual sunshine time 1 863.1–1 909.9 h. The pH in soil at 20 cm depth is 3.92–4.80. *D. dichotoma* samples were collected from LRM and NM of Longnan County in Jiangxi Province, China, respectively. The completely expended lamina was used for the following assay.

Isolation of chloroplasts was done according to Edwards *et al.* (1979) with little modification. The fresh lamina were thoroughly washed with de-ionized water, then ground in dark room at 4 °C for 20 s with blender in a medium containing 0.33 M sorbitol, 50 mM MES, 10 mM NaCl, 2 mM $MgCl_2$, 2 mM EDTA.Na_2, 0.5 mM KH_2PO_4, 2 mM Na-isoascorbate, and 0.20 % (m/m) bovine serum albumin (BSA) (pH 6.1). The slurry was filtered through 500, 195, and 20 μm nylon meshes and centrifuged at 300×*g* for 3 min. The pellets were suspended in the grinding medium and centrifuged at 5 000×*g* for 7 min to collect the chloroplasts. The isolated chloroplasts were washed with the grinding medium and re-suspended in the suspending buffer containing the same amounts as the grinding medium except replacing MES with 25 mM Hepes-NaOH (pH 7.6). The final chloroplast concentration was more than 1 kg(Chl) m^{-3} and stored in a refrigerator at –80 °C before use.

REEs determination in *D. dichotoma*: For each station, *D. dichotoma* samples were randomly collected, divided into root, stem, petiole, and lamina, and then mixed together. Samples were thoroughly washed with de-ionized water, then dried at 65 °C and ground to pass a 100-mesh sieve. These samples were dissolved by HNO_3/$HClO_4$/H_2O_2 ashing, and before determination by ICP-MS, 1 cm^3 of de-ionized water was added. The ICP-MS was conducted according to Jarvis (1992, 1997).

Fine location of REEs in the lamina cells: The electron microscopic analysis was done according to Shan *et al.* (2003). The intact lamina was thoroughly rinsed with tap water and distilled water, and then dried with filter paper. The samples (1 mm^3) were fixed in 3 % (m/v) glutaraldehyde and in 0.1 M phosphate buffer (pH 7.2) for 4 h, and washed with the phosphate buffer for 1 h. Then the samples were fixed with 1 % OsO_4 (pH 7.2) for 1 h, and rinsed with a buffer for three 10 min periods. After washing, the samples were dehydrated in a graded acetone and ethanol in series, and then embedded in Spurr's resin for 3 d. Dry sections (1–2 μm) were cut with a diamond knife using an ultramicrotome, and mounted on copper grids. Electron microscopic observation was made at 100 kV with a *Hitachi 800* electron microscope. Elemental X-ray microanalysis was performed using a *Philips EDAX 9100* microscope. A scanning transmission electron micrograph was first taken at 175 kV, followed by microanalysis for 100 s of 5.0 nm spots.

Photosynthetic electron transport activity of isolated chloroplasts from the fern was measured according to Leong and Anderson (1984) using a Clark-type electrode (*Hansatech*, UK) equipped with a circulating water jacket. The reaction mixture (1 cm^3) contained the chloroplasts equivalent to 50 mg(Chl) m^{-3}, 50 mM *Tricine*-NaOH (pH 7.5), 5 mM $MgCl_2$, 10 mM NaCl, 1 mM sodium ascorbate, and 0.4 M sucrose in the electron transport rate measurement of photosynthetic chain. The reaction mixture (1 cm^3) for the measurement of photosystem (PS) 2 electron transport activity contained 50 mM *Tricine*-NaOH (pH 7.0), 5 mM $MgCl_2$, 50 mM $CaCl_2$, 0.2 mM 2,6-dichloro-p-benzoquinone (DCBQ), 0.4 M sucrose, and chloroplasts equivalent to 50 mg(Chl) m^{-3}. The reaction mixture (1 cm^3) for the determination of PS1 electron transport rate contained 50 mM *Tricine*-NaOH (pH 7.5), 35 mM NaCl, 10 μM 3-(3,4-dichlorophenyl-1.1-dimethylurea) (DCMU), 1 mM sodium azide, 2 mM methyl viologen (MV), 1 mM sodium ascorbate, 0.2 mM 2.6-dichlorophenolindophenol (DCPIP), 0.4 M sucrose, and chloroplasts equivalent to 50 mg(Chl) m^{-3}. The assay temperature was adjusted to 20 °C. The photon flux density was 1 000 μmol(photon) m^{-2} s^{-1}.

Chlorophyll (Chl) fluorescence emission spectra and fluorescence kinetics: The fluorescence emission spectra of the fern chloroplasts were measured with a *Hitachi*

F-4500 fluorescence spectrophotometer at 77 K. The reaction mixture contained the isolated fern chloroplasts equivalent to 10 mg(Chl) m^{-3} and 50 % (v/v) glycerol. Modulated Chl *a* fluorescence measurements were made in attached lamina in the pots with a *PAM-2000* portable fluorometer (*H. Walz*, Effeltrich. Germany) connected to a notebook computer with data acquisition software (*DA-2000, H. Walz*, Germany).

Results and discussion

REEs contents in different parts and fine location in lamina cell of *D. dichotoma*: The REEs contents in *D. dichotoma* from LRM and NM areas were in sequence lamina>root>soil>stem>petiole (Table 1). Hence the REEs were absorbed by root and then transported through stem and petiole to plant lamina. The REEs contents were 2 648 and 1 494 mg kg^{-1}(d.m.) in the lamina of *D. dichotoma* from LRM and NM areas, respectively, thus the REEs content of LRM area was higher than that in NM plants by 77.1 %. REEs of the highest content in the lamina were LREEs La, Ce, and Nd. In the lamina of the plants from NM area, the percentages of these three kinds of REEs to total REEs contents were 30.7, 30.2, and 22.9 %, while in the lamina of the plant from the LRM area, the respective values were 41.4, 17.4, and 21.8 %.

Table 1. REEs contents [mg kg^{-1}] in different parts of *D. dichotoma* from two localities. ΣREE, the sum of Ln+Y in lamina; LR/HR, LREEs (La~Eu)/HREEs (Gd~Lu). δCe, Ce anomaly value = $Ce_N/(La_N \times Pr_N)^{0.5}$; δEu, Eu anomaly value = $Eu_N/(Sm_N \times Gd_N)^{0.5}$; REEn, chondrite-normalized REE value. NM: non-mining area, LRM, light REEs mine.

REEs	NM Soil	Root	Stem	Petiole	Lamina	LRM Soil	Root	Stem	Petiole	Lamina
La	36.20	179.58	30.52	12.31	458.53	55.27	136.36	35.54	53.33	1095.80
Ce	81.00	80.91	20.87	9.20	451.98	160.53	165.33	25.37	29.19	461.40
Pr	8.05	46.88	5.05	2.15	94.64	12.26	38.79	6.59	7.84	155.62
Nd	28.05	196.69	18.01	7.86	342.59	47.56	170.32	24.59	26.85	577.94
Sm	1.95	42.40	2.70	1.22	45.08	10.13	39.60	4.42	3.71	89.77
Eu	0.74	6.25	0.41	0.17	5.36	2.43	6.44	0.66	0.46	10.84
Gd	4.35	43.15	3.08	1.33	36.42	11.67	45.26	5.38	3.49	74.79
Tb	0.64	6.11	0.37	0.17	3.23	1.83	7.10	0.73	0.37	8.08
Dy	3.37	32.56	2.02	0.80	10.38	11.08	41.75	3.83	1.60	32.24
Ho	0.63	5.99	0.38	0.12	1.51	2.31	8.44	0.78	0.25	5.07
Er	1.88	15.06	1.06	0.38	4.13	6.82	23.28	2.18	0.79	12.21
Tm	0.25	1.89	0.12	0.03	0.30	1.01	3.23	0.26	0.07	1.08
Yb	1.70	10.76	0.70	0.20	1.46	6.78	19.53	1.49	0.42	5.00
Lu	0.25	1.59	0.09	0.01	0.17	1.01	2.76	0.20	0.05	0.59
Y	18.40	181.70	13.28	4.80	38.68	53.33	218.25	25.59	8.24	118.34
ΣREEs	172.06	851.52	98.64	40.74	1494.45	330.68	926.43	137.6	136.66	2648.79
LR/HR	12.15	4.72	9.93	10.87	24.28	6.78	3.68	6.54	17.23	17.20
δCe	1.11	0.21	0.40	0.40	0.51	1.44	0.53	0.39	0.33	0.26
δEu	0.49	0.43	0.43	0.40	0.40	0.67	0.46	0.41	0.39	0.40

As determined by transmission electron microscopy (TEM) and energy-dispersive X-ray microanalysis (EDAX) (Fig. 1), REEs were clustered in cell wall, cytoplasm, and chloroplasts in epidermal and mesophyll cells in the NM and LRM plants. The REEs deposits were found in the chloroplasts of intact mesophyll cells, mainly in thylakoid grana. It was previously suggested that the REEs can not penetrate into cytoplasm so that they can be used as ion tracer (Hall *et al*. 1977, Block *et al*. 1998), while some researchers demonstrated that the REEs can penetrate into intact cell by a symplasm pathway (Shan *et al*. 2003) and ion channels (Peeters *et al*. 1989). Our results showed that a part of REEs was located at the grana area of the chloroplast thylakoid membranes in *D. dichotoma*.

The laminae of fixed area were cut and Table 2 shows the Chl (*a*+*b*) and β-carotene (β-car) contents and Chl *a/b* ratio in the same area of lamina. The Chl contents in lamina of *D. dichotoma* were greater in LRM than NM plants ($p<0.01$). The contents of β-Car were LRM>NM ($p<0.01$). The high amounts of β-Car may be associated with the adoption of *D. dichotoma* in REEs mine: β-Car could quench extra energy when photosynthesis membrane was damaged. The Chl *a/b* of LRM plants was changed little compared to NM, suggesting that the Chl composition remained stable.

Table 2. Photosynthetic pigment (Chl: chlorophyll, β-Car: β-carotene) contents [μmol m^{-2}] in the lamina of *D. dichotoma* in NM and LRM plants.

Place	Chl ($a+b$)	a/b	β-Car
NM	167.00±6.52	2.14±0.05	2.40±0.21
GX	218.30±10.64	2.11±0.03	13.98±0.95

Photosynthetic electron transport activities of the total chain and PS2 (Fig. 2) in chloroplasts from LRM area plants were higher than those in chloroplasts of NM area plants by about 34.9 and 252.9 % (p<0.01), respectively, in contrast with the PS1 activity that was higher by 16.8 % (p<0.01) in the chloroplasts from LRM plants than NM plants. Thus REEs stimulated PS2 more than PS1.

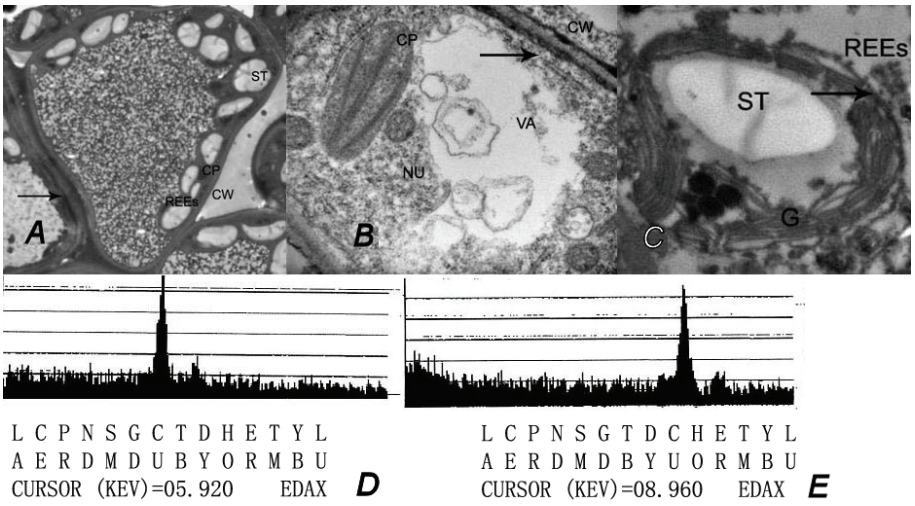

Fig. 1. The mesophyll cell (*A*) in lamina in LRM plants, epidermal cell (*B*) in lamina of NM plants, and chloroplast in LRM plants (*C*) with EDAX results (*D, E*). CP, chloroplast; CW, cell wall; G, grana; NU, nucleolus; ST, starch; REEs, REEs deposit; VA, vacuole.

Fluorescence emission spectrum and light-induced Chl *a* fluorescence kinetics: Two main fluorescence peaks were located at 684 (F_{684}) and 720 (F_{720}) nm (Fig. 3), the former being caused by PS2 and the later by PS1. The ratio of F_{684}/F_{720} reflects the distribution of excitation energy absorbed by Chl between the two photosystems. This ratio for the chloroplasts from plants of LRM area (1.89±0.04) was higher than that of the chloroplasts from NM area (1.77±0.05) by about 6.8 %. This indicates more excitation energy distributed to PS2 than PS1 in LRM plants. This may be one of the reasons for the increase in the electron transport activity of PS2 in these plants.

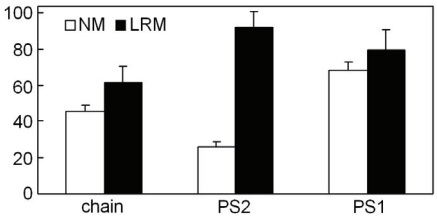

Fig. 2. The electron transport of photosynthetic chain [%] of photosystem 2, PS2 (oxygen evolution) and photosystem 1, PS1 (oxygen uptake) of chloroplasts. Means ± SD of three samples.

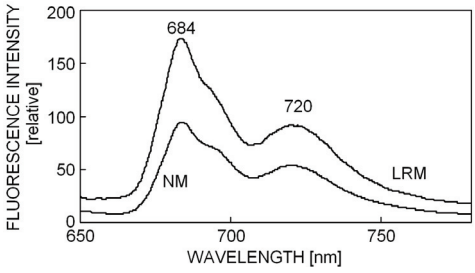

Fig. 3. The fluorescence emission spectrum at 77 K (excited at 436 nm) of chloroplasts from LRM and NM plants.

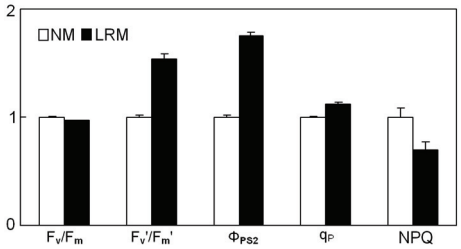

Fig. 4. The comparison of light induced Chl *a* fluorescence parameters in lamina from LRM and NM plants of *D. dichotoma*. Means ± SD of three samples.

Fig. 4 shows the light-induced Chl *a* fluorescence kinetic parameters (Genty *et al*. 1989) of the lamina from NM and LRM plants. Although the maximal efficiency of PS2 photochemistry (F_v/F_m) was only little different in the two plants, the actual photochemical efficiency of PS2 (Φ_{PS2}), the efficiency of excitation energy trapped by

open PS2 reaction centres in the light-adapted state (F_v'/F_m'), and photochemical quenching (q_P) increased by 54.8, 76.0, and 12.7 % ($p<0.01$), respectively. Meanwhile, the non-photochemical quenching (q_N) which reflects the process competing with PS2 photochemistry for absorbed excitation energy (Campbell *et al*. 1998) decreased by 30 % ($p<0.01$) in the lamina of LRM plants in contrast to NM plants. This was consistent with the content of β-Car. Our results show the efficiency of excitation energy trapped by PS2 reaction centre, the quantum yield of primary photochemical reaction, and the efficiency of photon energy utilization of PS2 are remarkably better in LRM plants of *D. dichotoma*.

Usually, the strategies plants use to cope with high concentrations of toxic metal were to deposit them (Küpper *et al*. 1999, 2001) and change their physiological characters. It suggests that the mechanisms of hyperaccumulation of REEs by *D. dichotoma* were to fix REEs in the cell wall, vacuoles, and chloroplasts and alter their physiological characters, such as use of β-Car to avoid the direct effect of high concentrations of REEs on PS2 photosynthetic characteristics.

References

Bayer, M.E., Bayer, M.H.: Lanthanide accumulation in the periplasmic space of *Escherichia coli* B. – J. Bacteriol. **173**: 141-149, 1991.

Block, B.M., Stacey, W.C., Jones, S.W.: Surface charge and lanthanum block of calcium current in bullfrog sympathetic neurons. – Biophys. J. **74**: 2278-2284, 1998.

Campbell, D., Hurry, V., Clarke, A., Gustafsson, P., Öquist, G.: Chlorophyll fluorescence analysis of cyanobacterial photosynthesis and acclimation. – Microbiol. mol. Biol. Rev. **62**: 667-683, 1998.

Chang, J.: [Effects of lanthanum on the permeability of root plasmalemma and the absorption and accumulation of nutrients in rice and wheat.] – Plant Physiol. Commun. **27**: 17-21, 1991. [In Chin.]

Chen, W.J., Gu, Y.H., Zhao, G.W., Tao, Y., Luo, J.P., Hu, T.D.: Effects of rare earth ions on activity of RuBPcase in tobacco. – Plant Sci. **152**: 145-151, 2000a.

Chu, H.Y., Li, Z.G., Xie, Z.B., Zhu, J.G., Cao, Z.H.: [Effects of lanthanum on the microflora of red soil.] – Environ. Sci. **21**(6): 28-31, 2000b. [In Chin.]

Chu, H.Y., Zhu, J.G., Xie, Z.B., Li, Z.G. Cao, Z.H.: [Effects of rare earth element lanthanum on the activities of invertase, catalase and dehydrogenase in red soil.] – Chin. environ. Sci. **21**(1): 77-80, 2001. [In Chin.]

Clijsters, H., van Assche, F.: Inhibition of photosynthesis by heavy metals. – Photosynth. Res. **7**: 31-40, 1985.

David, P., Karlish, S.J.D.: Characterization of lanthanides as competitors of Na^+ and K^+ in occlusion sites of renal (Na^+ K^+)-ATPase. – J. biol. Chem. **266**: 14896-14902, 1991.

Edwards, G.E., Lilley, R.M.C., Craig, S., Hatch, M.D.: Isolation of intact and functional chloroplasts from mesophyll and bundle sheath protoplasts of the C_4 plant *Panicum miliaceum*. – Plant Physiol. **63**: 821-827, 1979.

Genty, B., Briantais, J.-M., Baker, N.R.: The relationship between the quantum yield of photosynthetic electron transport and quenching of chlorophyll fluorescence. – Biochim. biophys. Acta **990**: 87-92, 1989.

Hall, A.E., Thomson, W.W., Asbell, C.W., Platt-Aloia, K., Leonard, R.T.: Stomatal response to humidity and lanthanum. – Physiol. Plant. **41**: 89-94, 1977.

Hong, F.S., Wei, Z.G., Tao, Y., Wan, S.K., Yang, Y.T., Cao, X.D., Zhao, G..W.: Distribution of rare earth elements and structure characterization of chlorophyll-lanthanum in a natural plant fern *Dicranopteris dichotoma*. – Acta bot. sin. **41**: 851-854, 1999.

Hu, X., Ding, Z., Wang, X., Chen, Y., Dai, L.: Effects of lanthanum and cerium on the vegetable growth of wheat (*Triticum aestivum* L.) seedlings. – Bull. environ. Contam. Toxicol. **69**: 727-733, 2002.

Ichihashi, H., Hand, M., Tatsukawa, R.: Rare earth elements (REEs) in naturally grown plants in relation to their variation in soils. – Environ. Pollut. **76**: 157-162, 1992.

Jarvis, I.: Sample preparation for ICP-MS. – In: Jarvis, K.E., Gray, A.L., Houk, R.S. (ed.): Handbook of Inductively Coupled Plasma Mass Spectrometry. Pp. 172-224. Blackie, Glasgow 1992.

Jarvis, K.E.: Inductively coupled plasma-mass spectrometry (ICP-MS). – In Gill, R. (ed.): Modern Analytical Geochemistry. Pp. 171-187. Longman Singapore Publ., Singapore 1997.

Küpper, K., Lombi, E., Zhao, F.J., Wieshammer, G., McGrath, S.P.: Cellular compartmentation of nickel in the hyperaccumulator *Alyssum lesbiacum, Alyssum bertolonii* and *Thlaspi goesingense*. – J. exp. Bot. **52**: 2291-2300, 2001.

Küpper, K., Zhao, F.J., McGrath, S.P.: Cellular compartmentation of zinc in leaves of the hyperaccumulator *Thlaspi caerulescens*. – Plant Physiol. **119**: 305-311, 1999.

Lasat, M.M.: Phytoextraction of toxic metals. – J. environ. Qual. **31:** 109-120, 2002.

Leong, T.-Y., Anderson, J.M.: Adaptation of the thylakoid membranes of pea chloroplasts to light intensities. II. Regulation of electron transport capacities, electron carriers, coupling factor (CF_1) activity and rates of photosynthesis. – Photosynth. Res. **5**: 117-128, 1984.

Li, L.B., Meng, X.X., Wu, Z.M., Zhou, P.Z.: [Stimulating role of rare earth elements on cation-induced chlorophyll *a* fluorescence enhancement effect at room temperature.] – Bot. Res. **6**: 215-220, 1992. [In Chin.]

Li, L.B., Meng, X.X., Wu, Z.M., Zhou, P.Z.: [The effect of nitrogen nutrition on the efficiency of primary photosynthetic process of light conversion in cucumber plant at primary flowering stage.] – Bot. Res. **7**: 224-229, 1994. [In Chin.]

Merroun, M.L., Chekroun, K.B., Arias, J.M., Gonzalez-Munoz, M.T.: Lanthanum fixation by *Myxococcus xanthus*: cellular location and extracellular polysaccharide observation. – Chemosphere **52**: 113-120, 2003.

Peeters, G.A., Kohmoto, O., Barry, W.H.: Detection of La^{3+} influx in ventricular cells by indo-1 fluorescence. – Amer. J. Physiol. Cell Physiol. **256**: 351-357, 1989.

Peterson, T.A., Swanson, E.S., Hull, R.J.: Use of lanthanum to trace apoplastic solute transport in intact plants. – J. exp. Bot. **37**: 807-822, 1986.

Poovaiah, B.W., Leopold, C.: Effects of inorganic salts on tissue permeability. – Plant Physiol. **58**: 182-185, 1976.

Shan, X.Q., Wang, H.O., Zhang, S.Z., Zhou, H.F., Zheng, Y.,

Yu, H.,. Wen, B.: Accumulation and uptake of light rare earth elements in a hyperaccumulator *Dicropteris dichotoma.* – Plant Sci. **165**: 1343-1353, 2003.

Shen, B.L., Zhang, L.J.: [Effects of rare earth elements lanthanum on inner hormone of wheat seedling.] – Plant Physiol. Commun. **30**: 351-352, 1994. [In Chin.]

Wang, H.O., Shan, X.Q., Zhang, S.Z., Wen, B.: Preliminary characterization of a light rare earth element binding peptide of a natural perennial fern *Dicranopteris dichotoma*. – Anal. bioanal. Chem. **376**: 49-52, 2003a.

Wang, X.P., Shan, X.Q., Zhang, S.Z., Wen, B.: Distribution of rare earth elements among chloroplast components of hyperaccumulator *Dicranopteris dichotoma*. – Anal. bioanal. Chem. **376**: 913-917, 2003b.

Wang, Y.Q., Jiang, P., Guo, F..Q, Zhang, Z.Y., Sun, J.X., Xu, L., Cao, G.Y.: [REE bound DNA in natural plant.] – Sci. China B **29**: 373-378, 1999. [In Chin.]

Wang, Y.Q., Sun, J.X., Guo, F.Q., Chen, H.M., Xu, L., Cao, G.Y.: [REE bound polysaccarids in the leaves of *Dicranopteris dichotoma* by INAA.] – Sci. China (B). **27**: 517-521, 1997. [In Chin.]

Xu, G.X., Xiao, J.M. (ed.): New Frontiers in Rare Earth Science and Applications II. – Pp. 1504-1534. Science Press, Beijing 1985.

Yi, C., Huo, Q.H., Lu, J.F., Li, R.C., Wang, K.: The transport kinetics of lanthanide species in a single erythrocyte probed by confocal laser scanning microscopy. – J. biol. inorg. Chem. **4**: 447-456, 1999.

Zhou, S.G., Liu, M.: [X-ray energy spectra analysis of lanthanum in tissue cells of different parts of wheat plant.] – Acta bot. sin. **40**: 180-183, 1998. [In Chin.]