

## Distribution and Mobility of Mineral Elements in Cultured *Gentiana rigescens* and Soil

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### ABSTRACT

Distribution and mobility of nine mineral elements (K, Ca, Na, Mg, Fe, Zn, Cu, Se, and Cr) in cultured *Gentiana rigescens* and its root zone soil were determined by atomic absorption spectroscopy. *G. rigescens* materials were planted under tea and white papaya trees in Yun County and Yongde of Lincang area in Yunnan Province of China. The results showed that *G. rigescens* and its soil were high in K, Ca, Fe, and Mg. The concentrations of Na and Mg in the soil were significantly lower than that in the plants. However, the concentrations of Fe, Cu, Se, and Cr were significantly higher in soils than that in the plant parts. *G. rigescens* roots showed high accumulation ratios for Na, Mg, and K, but low accumulation ratios for Fe, Cu, Se, especially for Cr. Transfer coefficients indicated *G. rigescens* might be a good accumulator for Ca.

**Keywords:** Accumulation, Ecological planting, Atomic absorption spectroscopy, Transfer coefficient.

### INTRODUCTION

Gentian (the family Gentianaceae) is commonly used as a traditional Chinese medicinal material, possessing effects of heat-clearing and fire-purging, liver protecting, spleen reinforcing and antibacterial activity (Li *et al.*, 2009). In 'Chinese Pharmacopoeia' (The state Pharmacopoeia Commission, 2010), the dried roots and rhizomes of *Gentiana manshurica*, *G. scabra*, *G. triflora*, and *G. rigescens* were documented as the materials of gentian. Most of the first three species are distributed in the northeast of China, while *G. rigescens* is in the southwest, mainly in Yunnan, Guizhou, Sichuan, and Guangxi Provinces (He *et al.*, 1988). Importantly, *G. rigescens* is one of the genuine medicinal materials of Yunnan Province and is distributed in the areas of Lincang, Chuxiong, Diqing, Pu'er, Kunming and Honghe. Normally, it grows in the adret,

meadow and open forest with an elevation of 1,900-2,500 m (Li *et al.*, 2009). Primary active constituents in *G. rigescens* are gentiopicroside, swertiamarin, sweroside, and erythricine (Yang *et al.*, 2003). Modern pharmacological study suggests that gentiopicroside has significant hepatic and stomachic efficacy, and erythricine possesses effects of anti-inflammation, anti-hyperthyroidism, blood glucose raising, blood press reduction, and bacteriostasis. Clinically, gentian is used to cure many diseases, such as hepatic and biliary disorder, hypertension, dermatosis, acute pharyngitis and conjunctivitis (Zhang *et al.*, 1991; Liu *et al.*, 2004). In germ plasm resource of *G. rigescens* from different places of Yunnan, contents of gentiopicroside are higher than the specified value 1.5% (Li *et al.*, 2008; Chinese Pharmacopoeia, 2010; Lai *et al.*, 2010).

*G. rigescens* produced in Yunnan has attracted much attention because of its

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pleasing shape, high content of effective components, favorable clinical efficacy, and wide application in medicine industry. According to statistics, there are more than 20 pharmaceutical companies in Yunnan, which use *G. rigescens* as the main material to produce Chinese patent drugs and the demand of *G. rigescens* is nearly 1,000 ton per year (Li et al., 2009).

Before 2005, the market demand for *G. rigescens* mainly relied on wild resource in Yunnan. However, the *G. rigescens* resources declined sharply because of the extensive excavation. At present, the shortage of *G. rigescens* has become the bottleneck of the sustainable development of medicine enterprises. As a perennial medicinal plant, the medicative parts of *G. rigescens* are root and rhizome. Additionally, it is usually grown in open forest and grass slope together with deep root systems. Disorderly digging of *G. rigescens* brought about a series of environmental problems, such as mountain vegetation destruction, soil erosion, ecotope deterioration, and biodiversity reduction. Therefore, it is urgently needed to protect *G. rigescens* resources and realize its sustainable utilization.

Sustainable production systems have been proposed as a strategy to minimize environmental impacts caused by agriculture. Agricultural sustainability is based on conservation practices, as well as efficient use of natural resources (Ferreira and Martin-Didonet, 2012). The cultural technique of artificial domestication is one effective method to ensure the sustainable supply of *G. rigescens*. Since 2000, people in Yun County and Yongde of Lincang area in Yunnan have begun to culture *G. rigescens*. Presently, planting *G. rigescens* has become an important income source of local residents. At the same time, the two places are also important production bases for tea and white papaya. As *G. rigescens* adapts to a shade environment, planting *G. rigescens* together with tea and white papaya seems to be a good choice. Not only it saves land resources, but also protects the

ecological environment of mountainous area from damaging and meets the market demand for *G. rigescens*.

Plants are significant components of ecosystems for their transferring elements from abiotic into biotic environments. Mineral elements play an important role in the formation of active constituents in medicinal plants. However, some micronutrient (e.g., Cu, Cr, and Zn) may be toxic to both plants and animals at high concentration (Chojnacka et al., 2005). Bioavailability means the ability of an element to be transferred from the soil to a living organism (Kabata-Pendias and Pendias, 2001). Therefore, we investigated the mineral elements profile in *G. rigescens* planted under tea and white papaya trees in Yun County and Yongde of Lincang area in Yunnan Province, including distribution and mobility of nine mineral elements in the *G. rigescens* and its soil.

## MATERIALS AND METHODS

### Materials

*G. rigescens* and its soil samples were collected from 14 locations in Yun County and Yongde of LinCang area in Yunnan Province of China. From each site, 1-kg soil samples were gathered from 0-30 cm of the soil layer by the plum flower stationing method. The reference substances (standard solutions of elements 1,000  $\mu\text{g}\cdot\text{mL}^{-1}$ ) were provided by the National Research Center for Certified Reference Materials (NRCCRMS). The analytical reagents (nitric acid and muriatic acid) were from Beijing Beihua Fine Chemical Co., Ltd. The water was deionized water (18.25 M $\Omega$ .cm).

### Sample Preparation

Prior to analysis, whole plants were partitioned into roots, stems, leaves, and flowers. The plant samples were dried at 60°C until constant weight. A powdered

sample of  $0.5000 \pm 0.0005$  g was digested in moderately concentrated nitric acid (70%) for 12 h and, then, was heated until the reddish brown fumes disappeared. The residue was heated once again at  $550^\circ\text{C}$  for 4 hours, then was digested in  $1 \text{ mol}\cdot\text{L}^{-1}$  muriatic acid up to 25 mL for determination. A blank control group was treated in the same way. All the determinations were performed in quintuplicate (Jabeen *et al.*, 2010).

After natural drying for one week and picking out organic debris, soil samples were powdered through 80 meshes. A  $0.5000 \pm 0.0005$  g soil sample was introduced directly into microwave polytetrafluoroethylene vessels and subjected to a 10 ml  $\text{HNO}_3$ -5 ml  $\text{HClO}_4$ -1 ml HF digestion procedure (Gong *et al.*, 2007). The digests were heated until the color of sample changed to primrose yellow. The solution was then made up to 25 mL with  $1 \text{ mol L}^{-1}$  muriatic acid.

### Instruments

Soil sample was pretreated by microwave digestion system (DV-600, Shanghai Silong scientific instruments company). All mineral elements were determined by atomic absorption spectrometer (SOLAAR, Thermo Elemental, USA). The GBW07408 (GSS-8) loess standard reference material (National Analysis Center, Beijing, China) was applied. The obtained results were in good agreement with certified values as shown in Table 1.

### Statistical Analyses

Accumulation ratios of mineral elements were calculated in this paper as they could provide comparison between different types of plants and soils, which also provide further understanding of the relationship between mineral element concentrations in soils and plants. Accumulation ratios, defined as the ratios of element concentration in plant root/element concentration in soil (Chopin *et al.*, 2008), were determined. Transfer coefficient (TC) was also calculated because it could explain what proportion of the element concentration in the roots is available and transferred to plant aerial parts. Transfer coefficient was defined and calculated as element concentration in aerial part /element concentration in root; for example,  $TC_{leaves} = [M]_{leaves} / [M]_{root}$  (Chopin *et al.*, 2008). Statistical analyses were carried out using SPSS 17.0 (Chicago, IL, USA) for Windows.

## RESULTS AND DISCUSSION

### Mineral Element Contents in *G. rigescens* and Soils

The total average concentrations of mineral elements in the plants were in the order of K ( $3075.03 \text{ mg}\cdot\text{kg}^{-1}$ ); Ca ( $1952.52 \text{ mg}\cdot\text{kg}^{-1}$ ); Mg ( $1119.07 \text{ mg}\cdot\text{kg}^{-1}$ ); Fe ( $854.96 \text{ mg}\cdot\text{kg}^{-1}$ ); Na ( $285.61 \text{ mg}\cdot\text{kg}^{-1}$ ); Zn ( $55.71 \text{ mg}\cdot\text{kg}^{-1}$ ); Se ( $27.95 \text{ mg}\cdot\text{kg}^{-1}$ ); Cr ( $5.87$

**Table 1.** Determined and certified values of trace elements in GBW07408 (GSS-8) loess (n= 10).

Element	Certified value	Determined	Recovery (%)
K	$2.42 \pm 0.06 \%$	$2.40 \pm 0.05 \%$	99.2
Ca	$8.27 \pm 0.18 \%$	$8.26 \pm 0.16 \%$	99.9
Na	$1.72 \pm 0.06 \%$	$1.73 \pm 0.03 \%$	100.6
Mg	$2.38 \pm 0.10 \%$	$2.33 \pm 0.15 \%$	97.9
Fe	$1.22 \pm 0.07 \%$	$1.20 \pm 0.10 \%$	98.4
Cu	$24.3 \pm 1.8 \text{ mg}\cdot\text{kg}^{-1}$	$24.5 \pm 1.0 \text{ mg}\cdot\text{kg}^{-1}$	100.9
Zn	$68 \pm 6 \text{ mg}\cdot\text{kg}^{-1}$	$67 \pm 5 \text{ mg}\cdot\text{kg}^{-1}$	98.5
Se	$0.12 \pm 0.04 \text{ mg}\cdot\text{kg}^{-1}$	$0.11 \pm 0.06 \text{ mg}\cdot\text{kg}^{-1}$	91.7
Cr	$68 \pm 8 \text{ mg}\cdot\text{kg}^{-1}$	$69 \pm 4 \text{ mg}\cdot\text{kg}^{-1}$	101.5



mg·kg<sup>-1</sup>), Cu (3.04 mg·kg<sup>-1</sup>). However, they were different in the soil, ranking in the order of Fe (4919.19 mg·kg<sup>-1</sup>); K (2913.73 mg·kg<sup>-1</sup>); Ca (1929.45 mg·kg<sup>-1</sup>); Mg (572.87 mg·kg<sup>-1</sup>); Cr (108.70 mg·kg<sup>-1</sup>); Se (94.58 mg·kg<sup>-1</sup>); Na (46.86 mg·kg<sup>-1</sup>); Zn (46.51 mg·kg<sup>-1</sup>), Cu (8.95 mg·kg<sup>-1</sup>). Trace elements possess both curative and preventive effects on combating diseases. There is a wide scope to explore the preventive medicinal values of various trace elements such as Ca, Fe, and so on (Khan *et al.*, 2011).

Mineral elements accumulated in roots could be translocated to other plant organs (Kabata-Pendias and Pendias, 2001). In order to investigate transfer up to the food chain, mineral element concentrations in aerial parts of *G. rigescens* were determined. Concentrations of mineral elements in different parts (roots, stems, leaves, and flowers) of *G. rigescens* and in soils are shown in Table 2. Significant differences could be found in mineral element distribution within the plant parts and soils ( $p < 0.05$ ). The concentrations of Na and Mg in soils were significantly lower than that in the plants. However, the concentrations of Fe, Cu, Se, and Cr in soils were significantly higher than that in the plant parts. For Fe, Cu, Se, and Cr, there was no significant difference between the roots, stems, leaves and flowers. For K, Ca, Na, Mg, and Zn, the highest concentrations were all in the leaves of *G. rigescens*.

### Accumulation Ratios and Transfer Coefficients in *G. rigescens*

The accumulation ratios of *G. rigescens* roots for K, Ca, Na, Mg, Fe, Zn, Cu, Se, and Cr varied widely (Table 3). Roots accumulated the highest amount of Na with the ratio of 4.92. But for Cr, the accumulation ratio was the lowest with only 0.04. The order of decreasing accumulation ratios was:

Na>Mg>K>Zn>Ca>Cu>Fe>Se>Cr

Few studies report trace element concentrations in *Gentiana* (Dong *et al.*, 2006; Wen *et al.*, 2006; Wang, 2007; Yang *et al.*, 2008; Zhang *et al.*, 2010). However, as trace element uptake patterns are vegetation and organ specific (Chopin *et al.*, 2008), lack of data for *G. rigescens* roots in the literature does not allow comparison with *G. rigescens* from other places.

Mineral elements accumulated in roots could be transferred to other plant parts (Chopin *et al.*, 2008). Transfer coefficients of stems, leaves, and flowers for the same element were not significantly different (Table 3). However, the same part had different transfer coefficients for different elements. Leaves of *G. rigescens* had the highest Ca transfer coefficients with 3.12. For Fe, the stems of *G. rigescens* had the lowest transfer coefficient, i.e. 0.58. According to Baker (1981), plants were classified as accumulators (TC>1.5), indicators (0.5<TC<1.5), and excluders

**Table 2.** Concentrations (mg·kg<sup>-1</sup> dry matter) of mineral elements in different plant parts of *G. rigescens* and soils (mean±SE, n=14) <sup>a</sup>.

	K	Ca	Na	Mg	Fe	Zn	Cu	Se	Cr
Roots	2901.23 ±45.76 <sup>b</sup>	869.88 ±115.62 <sup>c</sup>	230.75 ±21.78 <sup>b</sup>	1008.33 ±36.19 <sup>b</sup>	1127.01 ±126.48 <sup>b</sup>	44.11 ±3.17 <sup>b</sup>	2.35 ±0.38 <sup>b</sup>	20.21 ±10.23 <sup>b</sup>	4.13 ±1.71 <sup>b</sup>
Stems	3023.11 ±73.71 <sup>ab</sup>	1977.93 ±205.01 <sup>ab</sup>	311.20 ±21.52 <sup>ab</sup>	1100.44 ±35.10 <sup>ab</sup>	656.97 ±101.37 <sup>c</sup>	52.59 ±3.67 <sup>b</sup>	3.79 ±0.97 <sup>b</sup>	18.52 ±7.55 <sup>b</sup>	3.77 ±1.49 <sup>b</sup>
Leaves	3211.39 ±58.96 <sup>a</sup>	2717.80 ±413.10 <sup>a</sup>	337.50 ±18.03 <sup>a</sup>	1194.80 ±31.67 <sup>a</sup>	879.96 ±78.44 <sup>bc</sup>	66.56 ±5.39 <sup>a</sup>	3.61 ±0.84 <sup>b</sup>	20.67 ±7.55 <sup>b</sup>	3.78 ±1.51 <sup>b</sup>
Flowers	3171.28 ±70.32 <sup>a</sup>	2266.90 ±375.47 <sup>ab</sup>	261.26 ±25.71 <sup>b</sup>	1176.83 ±32.23 <sup>a</sup>	748.28 ±76.31 <sup>c</sup>	59.88 ±6.55 <sup>ab</sup>	2.38 ±0.17 <sup>b</sup>	19.87 ±7.61 <sup>b</sup>	3.68 ±1.40 <sup>b</sup>
Soils	2913.73 ±100.37 <sup>b</sup>	1929.45 ±178.80 <sup>b</sup>	46.86 ±6.71 <sup>c</sup>	572.87 ±67.24 <sup>c</sup>	4919.19 ±35.59 <sup>a</sup>	46.51 ±5.08 <sup>b</sup>	8.95 ±1.28 <sup>a</sup>	94.58 ±1.32 <sup>a</sup>	108.70 ±9.08 <sup>a</sup>

<sup>a</sup> The data with different letters in the same line means significant difference at 0.05 level

**Table 3.** Accumulation (root/soil) ratios and transfer (aerial parts/soil) coefficients in *G. rigescens*.

	K	Ca	Na	Mg	Fe	Zn	Cu	Se	Cr
	Accumulation ratio								
Roots	1.00	0.45	4.92	1.76	0.23	0.95	0.26	0.21	0.04
	Transfer coefficient								
Stems	1.04	2.27	1.35	1.09	0.58	1.19	1.61	0.92	0.91
Leaves	1.11	3.12	1.46	1.18	0.78	1.51	1.53	1.02	0.91
Flowers	1.09	2.61	1.13	1.17	0.66	1.36	1.01	0.98	0.89

(TC<0.1) based on their transfer coefficient. Our results indicated that *G. rigescens* might be a good accumulator for Ca and an indicator for K, Na, Mg, Fe, Zn, Cu, Se, and Cr.

### CONCLUSIONS

The results indicated that *G. rigescens* and its soil were high in K, Ca, Fe, and Mg. The concentrations of Na and Mg in soils were significantly lower than that in the plants. But, the concentrations of Fe, Cu, Se, and Cr in soils were significantly higher than that in the plant parts. *G. rigescens* roots showed high accumulation ratios for Na, Mg, and K, but low accumulations ratios for Fe, Cu, Se, and especially for Cr. Transfer coefficients showed *G. rigescens* might be a good accumulator for Ca.

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## توزیع و تحرک عناصر معدنی در گیاه جنتیان *Gentiana rigescens* و خاک آن

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### چکیده

توزیع و حرکت نه عنصر معدنی شامل پتاسیم، کلسیم، سدیم، منیزیم، آهن، روی، مس، سلنیوم و کروم در گیاه کاشته شده جنتیان *Gentiana rigescens* و در خاک ریشه گاه آن با دستگاه جذب اتمی تعیین شد. گیاه مزبور زیر سایه بوته های چای و درختان پاپایای سفید در بخش های یون و یانگده از منطقه لینکانگ در استان یون-آن چین کاشته شدند. نتایج نشان داد که این گیاه و خاک آن حاوی مقدار زیادی  $Mg$ ،  $K$ ،  $Ca$ ،  $Fe$  بودند. غلظت  $Mg$  و  $Na$  در خاک به طور معنی داری کمتر از غلظت در گیاه بود. ولی، غلظت  $Cu$ ،  $Fe$ ،  $Se$  و  $Cr$  در خاک به طور معنی داری از غلظت آنها در اندام گیاه بیشتر بود. عناصر سدیم، منیزیم و پتاسیم در ریشه جنتیان نسبت انباشت زیادی نشان دادند ولی نسبت انباشت آهن، مس، سلنیوم و کروم پایین بود. بر اساس ضریب انتقال می توان گفت که جنتیان انباشگر خوبی برای کلسیم است.