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Study on Mineral Elements in *Salvia roborwskii* from the Qinghai-Tibet Plateau

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Abstract The contents of trace elements, including aluminum, barium, beryllium, calcium, cobaltium, chromium, copper, iron, potassium, lanthanum, lithium, magnesium, manganese, sodium, nickel, phosphorus, lead, sulfur, silicon, titanium, vanadium, and zinc, were determined by ICP-AES. The mean concentrations of the 22 elements were as follows: K>Ca>Mg>Fe>Al>Na>S>P>Ba>Ti>Mn>Zn>Sr>Cu>Ni=Cr>Pb>V>Li=La>Co>Be. Principal components analysis of SPSS was applied to study the characteristic elements in *Salvia roborwskii*. Five principal components which accounted for 89.288% of the total variance were extracted from the original data. The first factor accounted for 56.401% of the total variance, which meant that aluminum, barium, beryllium, calcium, and cobaltium were the characteristic elements in *S. roborwskii*. This useful new method was used to evaluate the quality of *S. roborwskii*, and to provide the scientific foundation for its utilization and further research.

Keywords *Salvia roborwskii* · Mineral element · ICP-AES · Principal components analysis

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Introduction

The genus *Salvia* is one of the largest and most important members of the family Lamiaceae with over 1,000 species throughout the world. It is widely distributed in tropical, subtropical, and temperate regions. The fleshy root of *Salvia roborwskii* is often used as a substitute for *Salvia miltiorrhiza* Bunge (Chinese name "Dan shen"). *S. roborwskii*, known as "ji zi ga bao" in traditional Tibetan folk medicine, is a famous annual or biennial herbal medicine, distributed in Gansu, Sichuan, Xizang, Yunnan, and Qinghai in China, especially in the Qinghai-Tibet Plateau. It grows in meadows, forest edges, and furrow valleys under forests at altitudes of 2,500 to 4,200 m [1]. *S. roborwskii* is generally used to remove liver inflammation for improving eyesight and alleviating pain [2].

Trace elements are an important part of traditional Chinese medicine and medicinal properties, its features are small number of large function, and play a key regulatory role in the activity of many biological molecules [3]. Because it contains the same growth genes, the regulations of distribution and content of trace elements generally could be found in the same kind of medicine, it can be used to distinguish Chinese medicine [4]. In this study we evaluated 22 trace elements in *S. roborwskii* collected from different areas and altitudes in the Qinghai-Tibet Plateau.

Materials and Methods

Materials

S. roborwskii was collected in August 2009 from the Qinghai Plateau at altitudes ranging from 2,500 to 4,300 m at 20 different locations (Table 1). At each site, the individual plants

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Table 1 Information on materials

Sample	Sites	Altitude (m)	Latitude	Longitude
1	Heiquanshuiku, Menyuan	2,836	37°12′04″	101°32′22″
2	Dabanshan A, Menyuan	3,000	37°15′53″	101°24′58″
3	Dabanshan B, Menyuan	3,080	37°24′52″	101°24′26″
4	Xianchen, Menyuan	2,865	37°22′35″	101°33′40″
5	Dunchuan, Menyuan	2,672	37°19′27″	101°49′33″
6	Xianmilinchang, Menyuan	2,710	37°17′05″	101°57′55″
7	Xianmidianzhan, Menyuan	2,728	37°13′58″	102°00′58″
8	Haomen, Menyuan	2,600	37°10′10″	102°08′59″
9	Shierpan mountain A, Huzhu	2,581	37°00′56″	102°17′53″
10	Shierpan mountain B, Huzhu	2,666	37°01′55″	102°14′54″
11	Shierpan mountain C, Huzhu	3,095	37°01′43″	102°14′20″
12	Bomuxia, Huzhu	3,030	37°00′00″	102°06′26″
13	Manmenxia, Huzhu	2,800	36°57′57″	101°53′23″
14	Hongtu mountain, Yushu	4,289	33°07′53″	99°39′41″
15	Lebagou, Yushu	3,541	32°56′10″	97°15′08″
16	Wenchenggongzhumiaogou, Yushu	3,960	32°53′51″	97°04′25″
17	Batangreruigou, Yushu	3,840	32°51′53″	97°05′07″
18	Ziqionggou, Yushu	3,835	32°52′00″	97°04′56″
19	Laji mountain, Guide	3,189	36°18′06″	101°33′25″
20	Riyue mountain, Huangyuan	3,477	36°26′27″	101°05′27″

were picked randomly and then later mixed. The roots were lightly washed in the field and again in the laboratory with bidistilled water in order to remove soil and avoid any surface contamination. These were then dried at room temperature and crushed to a homogeneous fine powder (100 mesh). For the analysis of 22 elements (Mg, Ca, V, Mn, Fe, Co, Ni, Cu, Zn, Ba, Sr, Be, Pb, S, P, Al, Li, La, Na, K, Cr, and Ti), an IRIS INTREPID ICP-AES (Thermo Electron Corporation, USA) was used. Nitric acid (HNO_3) and hydrochloric acid ($HClO_4$) were analytical grade quality. All solutions and dilutions were prepared with double-distilled deionized water. Working standard solutions were prepared from mixed standards. The single standard solution of Ti (100 mg/L) was purchased from Accustandard Company, USA.

Sample Preparation

Accurately weighed root samples (0.300 g each) and 10 mL of HNO_3 were mixed, then 2.5 mL of $HClO_4$ was added, and the samples were heated on a hot plate at 150–200°C for 3 h. Samples were allowed to cool and then transferred to 25-mL volumetric flasks for analysis.

Instrument Working Conditions

See Table 2 for the instrument working conditions.

Statistical Analysis

Principal components analysis (PCA) and cluster analysis were used to analyze the data on mass mineral elements after standard processing using factor analysis in SPSS15.0, and the international application procedure DECORANA of PC-ORD software was used for detrended correspondence analysis (DCA).

Results and Discussion

The concentrations of 22 minerals in *S. roborwskii* are shown in Table 3. Ca and K had a large loading variable and are multifunctional nutrients that form an essential part of many important enzymes [5]. Cu is essential for a variety of biochemical processes and is needed for certain critical

Table 2 Working conditions for ICP-AES

Radiofrequency power (W)	Cooling gas consumption (L/min)	Auxiliary gas consumption (L/min)	Atomization pressure (psi)	Pump speed (rpm)
1,150	15	0.5	26	100

Table 3 Results of 22 mineral elements in *S. roborwskii* (in micrograms per gram)

No.	Al	Ba	Be	Ca	Co	Cr	Cu	Fe	K	La	Li	Mg	Mn	Na	Ni	P	Pb	S	Sr	Ti	V	Zn
1	1.2805	0.2143	T	14.7863	0.0014	0.0093	0.0231	2.2362	17.6770	0.0026	0.0029	2.1536	0.0406	0.2392	0.0073	1.5391	0.0105	1.4031	0.0476	0.0719	0.0024	0.0657
2	3.4194	0.2760	0.0002	18.5230	0.0018	0.0125	0.0917	4.4802	23.1270	0.0040	0.0048	5.0903	0.0975	2.5150	0.0105	0.8766	0.0117	1.0146	0.0739	0.1225	0.0081	0.0616
3	2.2372	0.0968	0.0001	12.8275	0.0016	0.0098	0.0190	3.5001	18.4266	0.0027	0.0030	3.9444	0.0567	0.6886	0.0058	0.6048	0.0071	1.2007	0.0412	0.0848	0.0043	0.0579
4	1.1932	0.0957	T	10.4001	0.0009	0.0077	0.0280	1.7575	14.5571	0.0015	0.0020	2.9110	0.0308	1.3947	0.0107	1.9397	0.0078	1.0776	0.0769	0.0430	0.0021	0.0363
5	2.1504	0.0574	0.0001	15.1614	0.0012	0.0099	0.0276	3.1985	15.3101	0.0025	0.0026	5.2492	0.0606	1.1686	0.0099	0.8790	0.0040	1.1748	0.0938	0.0852	0.0035	0.0502
6	2.1750	0.1500	0.0001	15.2588	0.0015	0.0103	0.0261	3.4714	20.5751	0.0025	0.0031	3.0653	0.0528	0.2880	0.0110	1.9667	0.0075	1.6000	0.0673	0.0577	0.0044	0.0793
7	4.3941	0.0957	0.0002	22.5069	0.0023	0.0147	0.0236	6.1981	12.5389	0.0046	0.0036	4.6265	0.1311	0.6095	0.0124	0.7267	0.0100	1.5431	0.0716	0.1559	0.0078	0.0531
8	4.6346	0.1528	0.0003	21.7314	0.0019	0.0140	0.0319	6.2155	15.7973	0.0058	0.0053	4.9007	0.1297	0.4078	0.0123	1.4046	0.0133	1.8816	0.0901	0.1846	0.0080	0.0733
9	1.6455	0.0625	0.0002	13.0733	0.0008	0.0081	0.0204	3.0105	14.4121	0.0022	0.0013	2.3384	0.0513	0.3596	0.0084	0.4802	0.0106	1.0149	0.0501	0.0637	0.0032	0.0702
10	3.8815	0.1238	0.0002	17.7541	0.0012	0.0119	0.0279	5.4864	17.6714	0.0040	0.0026	3.6371	0.1008	0.2338	0.0103	2.9524	0.0138	2.5317	0.0461	0.1125	0.0064	0.1294
11	12.4997	0.1851	0.0007	22.1478	0.0052	0.0297	0.0540	16.1349	12.7486	0.0128	0.0099	5.7247	0.2829	0.3478	0.0251	1.4557	0.0185	1.8434	0.0745	0.3275	0.0210	0.1361
12	1.4105	0.1704	0.0001	15.0683	0.0012	0.0093	0.0695	1.9421	17.7494	0.0020	0.0026	4.0758	0.0617	7.9469	0.0065	0.5509	0.0036	1.4074	0.1000	0.0532	0.0031	0.0537
13	2.1228	0.1100	0.0001	14.7155	0.0011	0.0086	0.1171	2.5686	23.0571	0.0024	0.0023	4.8835	0.0643	3.0345	0.0273	1.6197	0.0049	0.9155	0.0924	0.0837	0.0051	0.0650
14	0.5995	0.1464	T	9.3131	0.0008	0.0070	0.0661	1.0856	19.9021	0.0009	0.0026	2.4265	0.0402	0.4645	0.0056	1.0788	0.0040	0.7209	0.0377	0.0221	0.0015	0.0623
15	0.6827	0.1163	0.0001	9.1771	0.0007	0.0070	0.1238	1.0712	16.7350	0.0006	0.0016	2.6544	0.0336	1.9321	0.0079	0.2849	0.0022	0.5523	0.0855	0.0233	0.0020	0.0504
16	1.0176	0.1318	T	11.6990	0.0008	0.0083	0.0910	1.4815	28.0687	0.0014	0.0009	4.1020	0.0513	0.8888	0.0071	1.0799	0.0054	0.9269	0.0336	0.0450	0.0027	0.0876
17	1.7433	0.4580	0.0002	14.3735	0.0015	0.0092	0.1113	2.7275	29.5830	0.0021	0.0022	2.3232	0.0776	0.4086	0.0089	1.1473	0.0059	0.9161	0.0557	0.0349	0.0038	0.0985
18	0.9245	0.1578	T	11.3438	0.0007	0.0064	0.0476	1.2443	25.6852	0.0020	0.0029	5.2086	0.0580	2.8668	0.0052	1.4120	0.0084	1.0299	0.0421	0.0361	0.0027	0.0847
19	1.5788	0.7431	0.0001	14.2817	0.0010	0.0078	0.0375	2.1464	25.2638	0.0017	0.0025	3.9507	0.0761	1.6262	0.0048	1.2931	0.0184	1.6810	0.0509	0.0539	0.0029	0.0580
20	2.9945	0.1458	0.0002	14.7824	0.0012	0.0113	0.0348	4.8533	22.4016	0.0038	0.0037	4.7738	0.0723	1.0521	0.0158	1.8333	0.0121	1.6417	0.0718	0.0846	0.0058	0.0883
	2.6293	0.1845	0.0001	14.9463	0.0014	0.0106	0.0536	3.7405	19.5644	0.0031	0.0031	3.9020	0.0785	1.4237	0.0106	1.2563	0.0090	1.3039	0.0652	0.0873	0.0050	0.0731
	2.6094	0.1578	0.0002	3.9335	0.0010	0.0050	0.0352	3.3360	5.0062	0.0026	0.0019	1.1484	0.0558	1.7740	0.0060	0.6320	0.0047	0.4715	0.0208	0.0708	0.0043	0.0255

T (trace): relative content <0.0001 $\mu g\ g^{-1}$

enzymes to function in the body [6]. Cu is also involved in nervous system function, in maintaining the balance of other useful metals in the body such as Zn and Mo, and is necessary for normal function of the immune system. Mg is not only an important electrolyte but is also responsible for correct nerve and muscle function. It also acts as a cofactor in more than 300 metabolic reactions [7]. Zn is a component of more than 270 enzymes, and its deficiency in an organism is accompanied by multisystem dysfunction [8]. At the same time, heavy metal and pesticide residues in medicinal materials have become a problem worldwide in recent years. Due to changes in the population's health behavior, the hazards of heavy metals are now well known, and it is necessary to carry out research into the contents of heavy metal and pesticide residues in traditional medicine.

In this study, the mean values of the 22 elements in *S. roborwskii* were as follows: K>Ca>Mg>Fe>Al>Na>S>P>Ba>Ti>Mn>Zn>Sr>Cu>Ni=Cr>Pb>V>Li=La>Co>Be. Pb, V, Li, La, Co, and Be were found in trace amounts (<0.0100 μg g^{-1}); the concentrations of K, Ca, Mg, Fe, Al, Na, S and P were much higher (>1.000 μg g^{-1}), especially Ca and K (>10 μg g^{-1}). According to the regulations to limit heavy metal residues in imported medicinal material by some countries and organizations, the main harmful elements which must be controlled are Pb, Cd, As, Hg, and Cu [9]. With regard to these regulations, the concentrations of metal elements in *S. roborwskii* were far below the limits. It was shown that the residues of Cu and Pb were safe, and the residues of other elements were also low, such as Co, Cr, Mn, Ni, and V; therefore, further research into the exploitation and application of *S. roborwskii* should be carried out.

Principal Components Analysis

PCA can determine the complex relationships between different targets. A few indicators represent the information on many original targets and are the composite indices known as the main constituent elements of the indicators. The data on element content was tested using the relevant statistics, and all indicators were linearly correlated with a different target, which conformed to the PCA requirement.

The eigenvalues and the percentage of variance (in percent) of principal components in plant elements are presented in Table 4. Five principal components were selected with eigenvalues more than 1. The first five principal components represented 89.288% of all the variable information on plant mineral contents.

Another rotated component matrix of the first five principal components is shown in Table 5. In the first principal component, Al, Be, Ca, Co, Cr, Fe, La, Li, Mn, Ti, and V had high-positive correlations, the first principal component explained approximately 56.401% of the total variation. Therefore, Al, Be, Ca, Co, Cr, Fe, La, Li, Mn, Ti, and V were the main characteristic elements, and represented the majority of the mineral pattern seen in *S. roborwskii*.

Table 4 Eigenvalues and percentage of variance principal of plant elements

Component	Initial Eigenvalues		
	Total	Variance ratio (%)	Cumulative ratio (%)
1	12.408	56.401	56.401
2	2.678	12.171	68.572
3	1.928	8.766	77.338
4	1.373	6.240	83.578
5	1.256	5.710	89.288

The Relationship Between Element Contents and Latitude, Longitude, and Altitude of the Sampling Sites

The results of DCA showed that the two axes were highly positively correlated with latitude and longitude, but not altitude (Fig. 1). Ba and K had a close relationship with

Table 5 Loading matrix of the first five components in the rotated component matrix of plant elements

Element	Principal component				
	1	2	3	4	5
Al	0.980	0.098	0.139	0.015	−0.059
Ba	0.015	−0.036	−0.026	−0.028	0.933
Be	0.955	0.022	0.052	0.013	0.004
Ca	0.749	0.346	0.166	0.261	0.070
Co	0.970	0.057	−0.022	−0.014	−0.024
Cr	0.980	0.095	0.081	0.012	−0.071
Cu	−0.033	−0.915	−0.120	0.196	0.103
Fe	0.973	0.137	0.144	−0.025	−0.078
K	−0.351	−0.579	0.257	−0.080	0.614
La	0.973	0.132	0.149	0.021	−0.051
Li	0.929	0.107	0.051	0.094	0.034
Mg	0.520	0.012	0.181	0.561	0.060
Mn	0.977	0.062	0.099	0.043	0.078
Na	−0.214	−0.178	−0.172	0.780	0.124
Ni	0.623	−0.281	0.365	0.288	−0.337
P	0.035	0.176	0.934	−0.111	−0.003
Pb	0.590	0.458	0.295	−0.137	0.462
S	0.408	0.617	0.583	0.068	0.157
Sr	0.217	−0.014	−0.111	0.818	−0.310
Ti	0.954	0.201	0.095	0.090	−0.093
V	0.985	0.015	0.131	0.055	−0.034
Zn	0.569	−0.180	0.595	−0.348	0.127

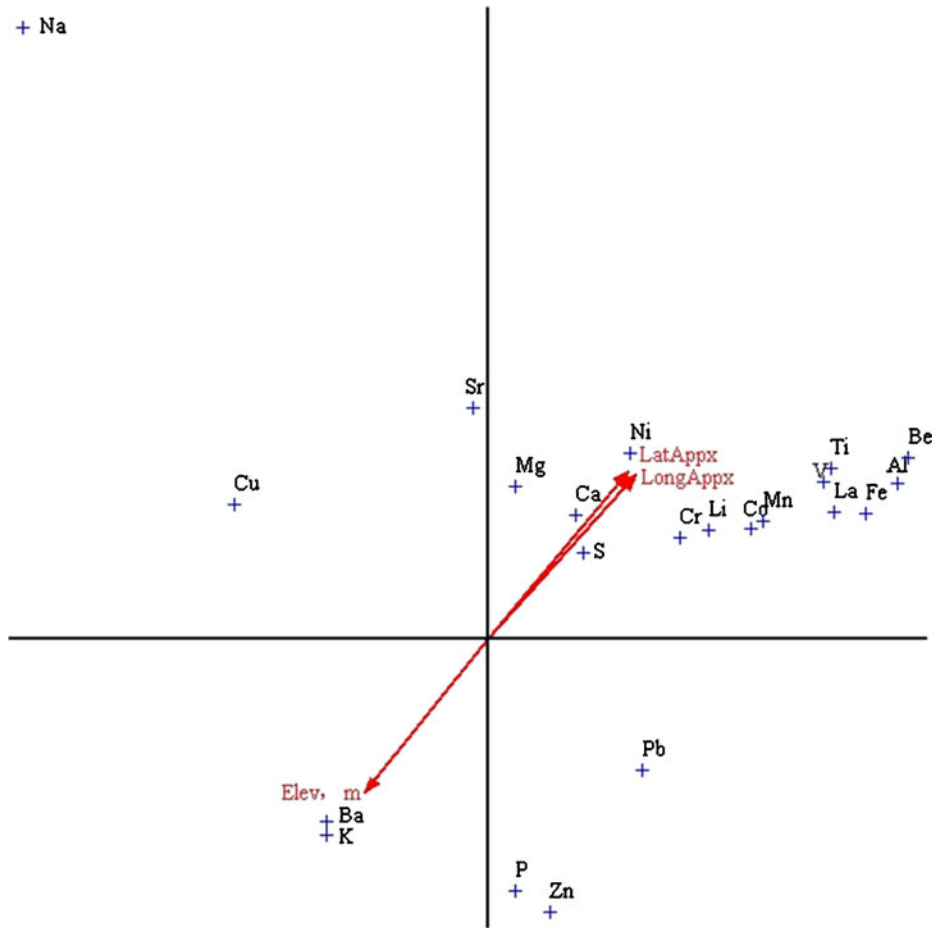

Fig. 1 The DCA ordination diagram of 22 mineral elements in *S. roborwskii* with latitude, longitude, and altitude

altitude. Ca, S, and Ni had a close relationship with latitude and longitude. The effect of latitude and longitude on the contents of elements was similar. Longitude and latitude are more correlated with the contents of most elements than altitude.

Conclusions

The concentrations of 22 minerals in *S. roborwskii* were determined. Principal components analysis showed that Al, Be, Ca, Co, Cr, Fe, La, Li, Mn, Ti, and V were the main characteristic elements in *S. roborwskii*. According to the regulations to limit heavy metal residues in imported medicinal materials by some countries and organizations, *S. roborwskii*, an important Tibetan herb, can be used safely. The effect of longitude and latitude on the contents of most elements was greater than that of altitude.

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