



Identification of geographical origins of raw American ginseng and tablets based on stable isotope ratios



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ABSTRACT

Background: American ginseng (*Panax quinquefolius*) is a medicine food homology plant, whose origin determines the medicinal and economical values. Therefore, a reliable method for the determination of its geographical origin must be established. An accurate, common and stable method to identify geographical origins of raw American ginseng and tablets was established by isotope ratio mass spectrometry (IRMS). 53 samples from 5 origins were collected and analyzed for isotope ratios of the elements C, H, O and N. The result showed that $\delta^2\text{H}$, $\delta^{18}\text{O}$ and $\delta^{15}\text{N}$ values were different among each geographical origin. The $\delta^2\text{H}$, $\delta^{18}\text{O}$ and $\delta^{15}\text{N}$ values were also used to establish discrimination model of geographical origin. According to verification test, the discrimination model of geographical origin was accurate and stable. The $\delta^2\text{H}$, $\delta^{18}\text{O}$ and $\delta^{15}\text{N}$ values were also found no significant difference between raw American ginseng and its tablet, so the discrimination model of geographical origin could also be used to discriminate the geographical origin of American ginseng tablet. In conclusion, the $\delta^2\text{H}$, $\delta^{18}\text{O}$ and $\delta^{15}\text{N}$ values can be used to discriminate the geographical origin of raw American ginseng and its tablet.

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1. Introduction

American ginseng is medicine food homology plant, the healthy security, richly containing the vitamin, nutrient and so on. American ginseng can boost one's energy and slow the aging process [1], help fight cancer [2], enhance the immune system, and improve cardio-cerebrovascular conditions; thus, it is also called "Green Gold". American ginseng, originally from the USA and Canada, was successfully introduced into China in 1975. The current main producing areas are USA, Canada, Shandong province, Jilin province and Beijing. The effectiveness and price of American ginseng from different origins are obviously different. Due to the difference in price, American ginseng is occasionally deplorably shoddy and passing off the false as genuine in market. So it is necessary to discriminate the origins of raw American ginseng and its tablet.

Most reports about discriminating geographical origins of traditional Chinese medicine mainly focused on chemical composition and shape [3,4]. To the best of our knowledge, stable isotope ratios are rarely proposed for discriminating the geographical origins of traditional Chinese medicine (TCM). There are many successful

examples about discriminating the geographical origins by using stable isotope ratio both at home and abroad. In recent years, the application of stable isotope ratios to discriminate geographical origins, especially in rice [5], orange juices [6], milk and milk products [7–9], meat [10,11], and honey [12], has made significant progress. The above examples show that stable isotope ratios are associated with geographical environment. The technology is based on geochemical information, so this method can also be used to discriminate the origins of TCM. In other words, Isotope Ratio Mass Spectrometry (IRMS) is a potential method to discriminate geographical origins of TCM.

Our study aimed to establish a discriminant model of geographical origins of American ginseng and its tablet by using discriminant analysis. According to this model, the geographical origins of American ginseng plants and tablets can be easily distinguished. However, for a more definitive conclusion, a much larger number of samples will need to be investigated.

2. Materials and methods

2.1. Standard reference materials

SNOW (Standard Mean Ocean Water); PDB (Pee Dee Belemnite); Nitrogen.

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Table 1
 $\bar{x} \pm SD$, minimum and maximum values of $\delta^{13}C$, δ^2H , $\delta^{18}O$ and $\delta^{15}N$ values (‰ vs. V-PDB, V-SMOW and N_2 , respectively) of all geographical origins.

Origins	$\delta^{15}N$ values(‰)			$\delta^{18}O$ values(‰)			δ^2H values(‰)			$\delta^{13}C$ values(‰)		
	Maximum	Minimum	$\bar{x} \pm SD$	Maximum	Minimum	$\bar{x} \pm SD$	Maximum	Minimum	$\bar{x} \pm SD$	Maximum	Minimum	$\bar{x} \pm SD$
Shandong	1.28	-2.46	1.85 ± 1.13	30.5	23.2	23.09 ± 1.80	-50.49	-72.27	-66.58 ± 5.02	-25.19	-25.99	-25.90 ± 0.89
Beijing	2.41	-0.63	-0.49 ± 0.63	24.42	19.93	22.47 ± 4.12	-71.36	-89.95	-67.35 ± 7.10	-24.39	-28.52	-25.94 ± 1.48
Jilin	3.97	-0.23	1.23 ± 1.49	24.21	20.5	23.24 ± 1.34	-71.36	-90.6	-84.16 ± 4.1	-22.19	-27.74	-25.51 ± 0.83
Canada	2.04	-0.79	0.34 ± 1.22	27.94	18.41	25.14 ± 3.28	-53.9	-67.4	-76.59 ± 4.41	-23.03	-28.3	-26.88 ± 1.12
America	2.87	-2.19	-0.05 ± 1.83	25.41	18.3	21.81 ± 2.20	-43.41	-60.41	-51.25 ± 0.95	-22.91	-27.37	-26.90 ± 0.95

2.2. Plant materials

A total of 53 authentic American ginseng samples were obtained from reliable sources such as the national institute for the control of pharmaceutical and biological products, and Beijing Tongrentang drug store, taking into account the main areas of American ginseng. On the basis of the origin, we identified 5 classes of samples: China–Shandong (no. = 9), China–Beijing (no. = 11), Canada–Vancouver and Ontario (no. = 10), America–Wisconsin (no. = 12) and China–Jilin (no. = 11). The domestic American ginsengs were dug in the field and dried at 60 °C, then measured in grams with dirt removed. American ginsengs of Canada (Vancouver and Ontario) were purchased in Canada. Some American ginsengs of America were got from the national institute for the control of pharmaceutical and biological products; the others were purchased in Beijing Tongrentang drug store.;

The species was identified by Dr. Chunsheng Liu, Beijing University of Chinese Medicine, China. The American ginseng tablets were purchased in some other drug stores.

2.3. Apparatus and methods

The analytical instrument is composed by a Flash EA1112 elemental analyzer, operating in combustion and pyrolysis modes, coupled to a Delta Plus XL. The continuous flow interface was a ConFlo III interface. All the devices are from Thermo.

2.3.1. Carbon and nitrogen contents for isotope ratio analysis

In general, analyses were carried out with 0.1–0.5 mg American Ginseng powder for carbon and nitrogen content measurements, respectively. Determination of $^{13}C/^{12}C$ and $^{15}N/^{14}N$ values was performed in combustion mode with an oxidative reactor (reactor temperature: 1120 °C). Gases produced by the oxidation reactor changed into NO_2 and CO_2 in the reducing furnace; He was used as carrier gas at a flow rate of 230 ml min⁻¹; the reactor temperature was 650 °C. After separation and purification, TCD and an isotope ratio mass spectrometer were used to assess $^{13}C/^{12}C$ and $^{15}N/^{14}N$ ratios.

2.3.2. Hydrogen and oxygen contents for isotope ratio analysis

Analyses were generally carried out with 0.3 mg American Ginseng powder for hydrogen and oxygen content measurements, respectively. Determination of $^2H/^1H$ and $^{18}O/^{16}O$ values was performed in combustion mode with an oxidative reactor (reactor temperature: 1450 °C). Gases produced by the oxidation reactor changed into H_2 and CO in the reducing furnace; He was used as carrier gas at a flow rate of 125 ml/min; the reactor temperature was 650 °C. After separation and purification, an isotope ratio mass spectrometer was used to assess $^2H/^1H$ and $^{18}O/^{16}O$ values.

Stability checks of the used reference gases were continuously performed by measuring International Atomic Energy Agency standards with defined $^{13}C/^{12}C$, $^2H/^1H$, $^{18}O/^{16}O$ and $^{15}N/^{14}N$ ratios. Isotope ratios were expressed in per mil (‰) deviation relative to the V-PDB, V-SMOW and N_2 international standards. Values were calculated according to the following formula:

$$\delta(\text{‰}) = \left[\frac{(R_{\text{sample}} - R_{\text{standard}})}{R_{\text{standard}}} \right] \times 100$$

where R_{sample} and R_{standard} are the isotope ratios of the sample and standard materials, respectively.

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