The ginsenosides and carbohydrate profiles of ginseng cultivated under mountainous forest

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ABSTRACT

Background: Ginseng cultivated under mountainous forest, called "Lin-Xia-Shan-Shen" (LXSS) in China's Pharmacopoeia. In recent years, it has been quickly propelled to plant at a large scale. Objective: To study the profiles of ginsenosides and carbohydrate profiles of LXSS. Materials and Methods: The contents of ginsenosides and carbohydrates, such as soluble sugar, polysaccharide, pectin, and starch in LXSS, were determined. All the above components were profiled, and the correlations between them were analyzed. Results: The results indicated that the contents of total ginsenoside, protopanaxadiol, protopanaxatriol, Rg₁, Re, Rb₂, Rc, Rb₂, Rd, starch, and pectin were negatively correlated with the growing years within 17 years. Among them, the content of starch was positively correlated with that of pectin. The total ginsenosides was positively correlated with starch and pectin, which cannot be found in garden ginseng, maybe resulting of fertilizer and other manual intervention in process of cultivation of garden ginseng. Discussion and Conclusions: The accumulation of ginsenosides and carbohydrate, especially starch and pectin, was different in garden ginseng and LXSS. This research may provide the scientific basis for germplasm evaluation, the cultivation and utilization of ginseng cultivated under mountainous forest.



Key words: Carbohydrate, ginseng, ginsenoside, pectin, polysaccharide, starch

INTRODUCTION

Ginseng, the dried roots of Panax ginseng C.A. Meyer, has been held in high esteem in traditional Chinese medicine for 400 years. It belongs to the family of Araliaceae and is a well-known medicinal plant worldwide. Traditionally, only wild ginseng was applied clinically in Traditional Chinese Medicine(TCM) However as the extinction of wild resources, the garden ginseng emerged 400 years ago. Now garden ginseng is the preponderant resource of medicinal ginseng materials, but the development of garden ginseng cultivation accompanies the deforestation and severe damage to the ecological balance as well. In recent decades, the ginseng cultivation under mountain forest has been spread at a large scale so as to preserve forest and imitate the growing conditions of wild ginseng. It was formally called "Lin-Xia-Shan-Shen" (LXSS) in China's Pharmacopoeia and named consuetudinary "Zi-Hai," meaning that the LXSS was cultivated directly by seeds and disseminated everywhere and grew naturally. Since the

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formulation of natural forest protection policy in 1998, the cultivation of garden ginseng shrunken and the cultivation of LXSS increased rapidly. However, the chemical study was rarely concerned. Previously, the bioactivities and hemolytic interaction of ginsenosides were studied, [1-4] and the SOP for cultivation and fingerprint of LXSS were proposed.^[5,6] The monthly accumulating profile of ginsenosides in LXSS and the optimal harvest month of LXSS were discussed. [7] More recently, the ginsenosides in garden ginseng and LXSS was compared. [7] Ginsenosides and pectin are medicinal components, and the soluble sugar, starch, and polysaccharide are the reflection of germplasm, such as resistance, genetic character, etc. Content of starch is relevant to the texture of LXSS. In continuation, the ginsenosides and carbohydrates of LXSS were studied. This paper dealt with the determination and correlations of the aforementioned components of LXSS.

MATERIALS AND METHODS

Materials and reagents

Samples of LXSS were collected in the county of Kuandian, Liaoning province and were identified as *Panax ginseng* C. A. Meyer by Professor Ting-guo Kang and Bing Wang. The growing years of LXSS were determined by the number of

rhizome nodes together with the record of collection areas. Samples of Garden ginseng were purchased from Shenyang Ginseng & Antler Market. The voucher specimens were deposited in College of Pharmacy, Liaoning University of Traditional Chinese Medicine. Sulfuric acid, anthrone, ethyl acetate, acetic acid, sodium hydroxide, iodine, ethanol, anhydrous ethanol, potassium iodide, soluble starch, glucose, and ethanol were analytical grade and purchased from Tianjin Kermel Chemical Reagent Co., Ltd. Pectinase was analytical grade and purchased from Shanghai Hualanchem Co., Ltd. d-Galacturonic acid with a purity of 97% was purchased from Sigma Aldrich Fluka Co. Deionized water was supplied by Xinshengyuan Company.

Instruments

Agilent 1100 HPLC instrument (Agilent Technologies, Inc.), AT-130 column oven (Dalian Zhonghuida Scientific Instrument Co., Ltd.), FW80 high-speed grinder (Tianjin Taisite Instrument Co., Ltd.), HITACHI UV-3010 spectrophotometer (HITACHI Group Ltd.), Sartorius CP225 analytical balance (Sartorius Co., Ltd.), and FW80 high-speed grinder (Tianjin Taisite Instrument Co., Ltd.) were employed during the process of chemical analysis.

Sample preparation

Preparation of samples for ginsenosides analysis

Ginsenosides were extracted and analyzed by methods of Zhou.^[7] Then, 0.5 g of the powder of ginseng was weighed accurately into a 250 mL round-bottomed flask and refluxed by methanol two times, 1 h for each time. The solvent was evaporated and the residue was dissolved by methanol to a constant volume, and then the solution was filtered through a suitable membrane (0.45 µm) before injection. Contents of ginsenosides with authentic samples such as Rg₁, Re, Rf, Rb₁, Rc, Rb₂, and Rd were also dissolved in methanol and determined by an external standard method.

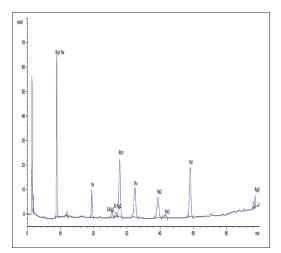


Figure 1: HPLC chromatogram of seven standard substances

The chemical analysis was achieved on an Agilent 1100 HPLC instrument with Agilent C_{18} column (150 × 4.6 mm, 5 µm), column temperature at 30 °C, and flow rate at 1.0 mL min⁻¹. The ginsenosides were separated in 70 min by the mobile phase consisted of acetonitrile (A) and 0.1% phosphoric acid (W) with the gradients as follows: 0-30 min, A:W from 19:81 to 29:71; 30-50 min, A:W from 29:71 to 32:68; 50–70 min, A:W from 32:68 to 51:49. While the determinations of ginsenosides Rg1 and Re were achieved by isocratic elution with A:W (20:80) within 30 min. Components with retention time between Re and Rf in the first condition were separated by gradient elution as follows: 0-30 min, A:W 20:80; 30-38 min, A:W from 20:80 to 21:79; 38-41 min, A:W from 21:79 to 21.5: 78.5; 41-57 min, A:B from 21.5:78.5 to 28:72 [Figures 1 and 2].

Preparation of samples for soluble sugars analysis

Soluble sugars were determined by the anthrone-H2SO4 method. [8] Fifty milligrams of the powdered sample were weighed accurately into a 10 mL centrifugal tube, and the mouth was covered with a plug. The samples were extracted with 10 mL of 80% ethanol at 80 °C in a water bath for 30 min with intermittent mixing. After cooling to room temperature, the samples were centrifuged at 4000g for 20 min and the supernatant was transferred into a 25 mL volumetric flask. The sediment was extracted and centrifuged as mentioned above again, then collected all the supernatant liquids and diluted it to 25.0 mL with water. Five milliliters of supernatant liquids were pipetted into a 50 mL volumetric flask and was made up to the mark line with water. Two milliliters of diluted solution was pipetted into a tube. Then, 0.5 mL of 2% anthrone-ethyl acetate solution was added and 5.0 mL of 98% H₂SO₄ was directed into the middle of the solution with care. The contents were mixed, and the tube was heated in a boiling water bath for 1 min. After cooling, the absorbance of the sample at

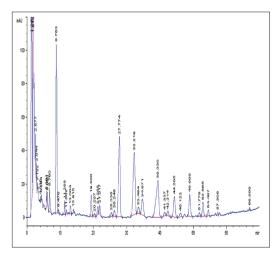


Figure 2: HPLC chromatogram of samples

630 nm was measured against a reagent blank as the reference. Standard curves were obtained by using d-glucose as a reference standard for colorimetric analyses. The soluble sugar concentration was cauculated by D-glucose.

Preparation of samples for total polysaccharide analysis

After washed with some 80% ethanol, the sediment obtained from centrifugation mentioned above was homogenized in 15.0 mL of 1 mol/L HCl, and extracted at 80 °C in a water bath for 1 h with vibration occasionally. After cooling to room temperature, 6 mL of 10% NaOH was added for neutralization. After centrifugation at 4000g for 10 min, the supernatant was obtained. When some supernatant was taken for reacting with iodine–potassiumiodide solution without the development of a blue color, 2.0 mL of supernatant was pipetted into a 50 mL volumetric flask and was make up to the mark with water. The following procedure was the same as that of measurement of soluble sugars content.

Preparation of samples for starch analysis

Starch was analyzed by using the iodine colorimetric method. [9] A 100 mg of the powdered sample was accurately weighed into a 10 mL centrifugal tube, 1 mL of anhydrous ethanol was added for wetting, then 9 mL of 1 mol/L NaOH was added, and covered the mouth with a plug. The samples were extracted with at 80 °C in a water bath for 10 min with intermittent mixing and then centrifuged at 4000g for 10 min after cooling to room temperature. Five milliliters of supernatant liquids were pipetted into a 25 mL volumetric flask and was make up to the mark with water. Some diluted solution was pipetted into a 100 mL volumetric flask, added with the corresponding volume of $0.09\,\mathrm{mol/L}\,\mathrm{NaOH}$ to make the total volume to $10\,\mathrm{mL}$. The reagents were added in the following order: 30 mL water, 1 mL of 1 mol/L acetic acid and 1 mL iodine solution and was made up to the mark with water. After 10 min, when the color was shown stably, the absorbance of the sample was measured at 580 nm against a reagent blank as the reference. Standard curves were obtained by using soluble starch as a reference standard for colorimetric analyses. Starch concentration was caculated by soluble starch.

Preparation of samples for pectin analysis

First, pectin was enzymatic hydrolyzed as Xu *et al.*^[10] descried and then measured by the 3,5-dinitrosalicylic acid (DNS) method of Miller.^[11] Five hundred grams of the powdered sample were accurately weighed into a 50 mL centrifugal tube, then operated as "measurement of soluble sugars content" mentioned above besides the volumes of 80% ethanol were 50 mL, and washed the sediment with some 80% ethanol. After evaporation, the sediment was enzymatic hydrolyzed with 50 mL of 0.1 M citric

acid—sodium citrate buffer (pH 4) and pectinase of 1200 u at 50 °C for 6 h. The blank was prepared as described above. Followed with filtration, the filtrate was transferred into a 50 mL volumetric flask and was made up to the mark with the buffer. One milliliter of the filtrate was pipetted into a 25 mL tube with a plug. Then, 3 mL DNS solution was added to each tube and boiled for 5 min. After cooling, the blank was added to make up to the mark, agitated, and the absorbance at 520 nm was measured. Standard curves were obtained by using d-galacturonic acid as a reference standard for colorimetric analyses. Pectin concentration was caculated by D-galacturonic acid.

Statistics

All values are expressed as mean \pm SD. The correlation analysis of data was achieved by SPSS.

RESULTS AND DISCUSSION

Method validation

The methods were validated by linearity, precision, repeatability, stability, and recovery, and all were up to the demands for HPLC and chromatometry determination.

Accumulation trend of ginsenosides in LXSS

The ginsenosides were determined by the procedure mentioned above. The following regularities could be concluded from the data as shown in Table 1, Figures 3, and 4.

- (1) The contents of total and protopanaxadiol (PPD) and protopanaxatriol (PPT) ginsenosides accumulated at the initial growing period (from the sixth to the eighth year) of LXSS, and reached a peak at the eighth year of growth, then fell off in the following years of growth. However, negative correlation between the content of total ginsenosides and growing years was figured out after SPSS analysis with the correlation coefficient (R value) of -0.956 (P < 0.01). The PPT and PPD ginsenosides and growing years were negatively correlated with the growing years with R values of -0.953 (P < 0.01) and -0.955 (P < 0.01). The ratio of PPT and PPD was positively correlated with the growing years with R value of 0.931 (P < 0.01).
- (2) At first glance, ginsenosides Rf, Rc, and Rb₂ showed the same accumulating trend with total ginsenosides [Table 2 and Figure 2], while the contents of Rg₁, Rb₁, and Rd fell off from the initial growing period of LXSS to the 17th year of growth. While the contents of Re fell off from the initial growing period of LXSS (from the sixth to the eighth year), then rose and fell off again in the following years of growth. But after SPSS analysis, all of them were negatively correlated with the growing years with R values of -0.961 (Rg₁, P < 0.01), -0.937 (Re, P < 0.01), -0.981 (Rb₁, P < 0.01), -0.847 (Rc, P < 0.05), -0.905 (Rb₂, P < 0.05),

Table 1: Contents of ginsenosides (mg/g) in LXSS with different growing years together with those of garden ginseng (n = 5)

Туре	Growing years	Rg₁	Re	Rf	Rb ₁	Rc	Rb_2	Rd	Total ginsenoside	PPT	PPD	PPT/ PPD	Rg₁/ Re
LXSS		3.61 ±	4.29 ±	1.32 ±	6.14 ±	5.38 ±	4.40 ±	1.84 ±		9.22 ±	17.76	0.52 ±	0.84 ±
	6	0.33	1.20	0.17	0.11	0.93	0.52	0.02	26.98 ± 0.05	0.18	± 2.37	0.03	0.04
		$3.58 \pm$	$3.95 \pm$	$1.78 \pm$	$5.98 \pm$	$5.96 \pm$	$4.72 \pm$	$1.77 \pm$		$9.31 \pm$	18.43	$0.51 \pm$	$0.91 \pm$
	8	0.07	0.21	0.04	0.26	1.16	0.38	0.06	27.74 ± 1.14	0.07	± 1.22	0.05	0.22
		$3.48 \pm$	$4.06 \pm$	1.13 ±	$5.78 \pm$	$5.57 \pm$	$4.07 \pm$	1.11 ±		$8.67 \pm$	16.53	$0.52 \pm$	$0.86 \pm$
	10	0.16	1.05	0.17	0.60	1.30	0.36	0.17	25.20 ± 2.33	0.64	± 2.10	0.15	0.16
		$3.45 \pm$	$3.40 \pm$	$1.04 \pm$	$5.17 \pm$	$4.95 \pm$	$3.46 \pm$	$1.09 \pm$		$7.89 \pm$	14.67	$0.54 \pm$	$1.01 \pm$
	12	0.36	0.41	0.09	0.21	0.38	0.27	0.13	22.56 ± 1.52	0.07	± 0.37	0.06	0.12
		$3.42 \pm$	$3.29 \pm$	$0.94 \pm$	$4.86 \pm$	$4.86 \pm$	$3.35 \pm$	$0.79 \pm$		$7.65 \pm$	13.86	$0.55 \pm$	$1.04 \pm$
	14	0.28	0.29	0.16	0.17	0.52	0.32	0.06	21.51 ± 0.08	0.31	± 0.22	0.21	0.29
		$3.39 \pm$	3.18 ±	$0.87 \pm$	$4.58 \pm$	$4.47 \pm$	$3.24 \pm$	$0.61 \pm$		$7.44 \pm$	12.90	$0.58 \pm$	1.07 ±
	17	0.40	1.07	0.14	0.26	0.17	0.07	0.10	20.34 ± 4.27	0.16	± 0.15	0.07	0.46
Garden		1.39 ±	2.20 ±	0.57 ±	2.25 ±	2.95 ±	3.09 ±	1.26 ±	10 = 1 : 0 10	4.16 ±	9.55 ±	0.44 ±	0.63 ±
ginseng	4	0.10	0.44	0.03	0.10	0.13	0.18	0.04	13.71 ± 0.10	0.07	2.08	0.12	0.02
		1.93 ±	2.46 ±	1.31 ±	3.40 ±	1.86 ±	1.74 ±	1.46 ±	4440.004	5.70 ±	8.46 ±	0.67 ±	0.78 ±
	4	0.07	0.17	0.02	0.36	0.27	0.53	.07	14.16 ± 0.21	0.13	0.10	0.06	0.16
	0	2.15 ±	2.99 ±	0.08 ±	3.01 ±	2.70 ±	2.79 ±	1.73 ±	40.47 + 0.07	5.94 ±	10.23	0.58 ±	0.72 ±
	6	0.16	0.39	0.05	0.42	0.38	0.36	0.06	16.17 ± 0.87	0.22	± 1.04	0.13	0.20
	4	1.81 ±	2.08 ± 0.08	0.73 ±	3.28 ± 0.51	2.83 ±	2.72 ± 0.42	1.33 ±	44.70 + 4.40	4.62 ±	10.16 ± 2.06	0.45 ±	0.87 ±
	4	0.13		0.27		0.16		0.15	14.78 ± 1.16	0.02		0.06	0.16
	4	1.91 ± 0.19	2.63 ± 0.33	1.02 ± 0.03	3.07 ± 0.29	2.55 ± 0.19	2.05 ± 0.26	1.21 ± 0.04	14.44 ± 0.07	5.56 ± 0.45	8.88 ± 0.13	0.63 ± 0.02	0.73 ± 0.07
	4	1.33 ±	1.84 ±	0.03 0.46 ±	1.98 ±	1.76 ±	1.83 ±	0.04 0.97 ±	14.44 ± 0.07	3.63 ±	6.54 ±	0.02 0.56 ±	0.07 0.72 ±
	4	0.25	0.02	0.46 ±	0.03	0.27	0.04	0.97 ±	10.17 ± 2.11	0.01	0.34 ± 0.22	0.08 0.08	0.72 ± 0.11
	7	2.21 ±	2.82 ±	0.55 ±	4.21 ±	4.01 ±	4.05 ±	1.88 ±	10.17 ± 2.11	5.58 ±	14.15	0.00 0.39 ±	0.78 ±
	6	0.13	2.62 ± 0.42	0.33 ±	4.21 ± 0.87	4.01 ±	4.03 ± 0.62	0.26	19.73 ± 1.18	0.17	± 0.17	0.39 ±	0.78 ± 0.19
	J	1.77 +	3.10 ±	1.23 ±	2.57 +	5.23 ±	2.67 ±	0.20 0.92 ±	10.70 ± 1.10	6.10 ±	11.39	0.54 ±	0.13 0.57 ±
	6	0.05	0.28	0.06	0.05	0.62	0.06	0.92 ±	17.49 ± 0.53	0.10 ±	± 1.26	0.15	0.04

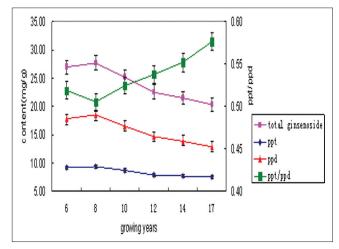


Figure 3: The accumulation trend of total ginsenoside, ppd, ppt and ppt/ppd of LXSS

and -0.957 (Rd, P < 0.01) while the content of Rf was not correlated with the growing years with R values of -0.772 (P > 0.05). The ratio of Rg₁ and Re was positively correlated with the growing years with a R value of 0.919 (P < 0.05).

Accumulation trend of carbohydrate in LXSS

The soluble sugar, starch, total polysaccharide, and pectin were determined by the procedure mentioned above. The

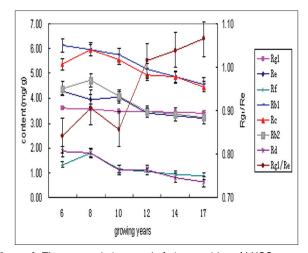


Figure 4: The accumulation trend of ginsenosides of LXSS

following regularities could be concluded from the data as shown in Table 2 and Figure 5.

(1) The content of soluble sugar accumulated at the initial growing period (from the sixth to the twelfth year) of LXSS, and reached a peak at the 12th year of growth, then fell off in the following years of growth. While the content of starch increased, reached the highest level

Table 2: Contents of carbohydrate (%) in LXSS with different growing years together with those of	
garden ginseng (n = 5)	

Туре	Growing years	Soluble sugar	Starch	Polysaccharide	Pectin
LXSS	6	26.62 ± 0.27	16.93 ± 0.21	41.45 ± 0.61	13.82 ± 0.15
	8	30.18 ± 0.32	22.24 ± 0.20	44.13 ± 0.45	14.36 ± 0.05
	10	40.34 ± 0.80	18.18 ± 0.18	48.10 ± 0.79	13.94 ± 0.08
	12	45.52 ± 0.78	16.16 ± 0.15	45.97 ± 1.53	13.11 ± 0.10
	14	34.99 ± 0.42	9.32 ± 0.08	43.05 ± 0.40	12.95 ± 0.14
	17	28.89 ± 0.24	3.44 ± 0.02	38.95 ± 3.19	12.70 ± 0.11
Garden	4	17.84 ± 0.17	38.92 ± 2.24	53.41 ± 3.04	11.36 ± 1.06
ginseng	4	18.45 ± 0.13	35.1 ± 1.33	55.29 ± 1.16	10.29 ± 1.04
	4	12.65 ± 0.08	43.82 ± 2.60	49.57 ± 0.82	12.92 ± 2.02
	4	7.67 ± 0.03	33.28 ± 0.22	69.47 ± 1.83	9.29 ± 0.07
	4	14.3 ± 0.11	28.16 ± 0.37	61.14 ± 1.05	11.14 ± 0.13
	6	32.56 ± 0.40	15.84 ± 0.07	25.13 ± 1.37	6.71 ± 0.01
	6	50.1 ± 3.62	39.31 ± 2.61	56.72 ± 3.92	12.69 ± 1.16
	6	54.43 ± 3.66	27.79 ± 0.14	39.06 ± 1.47	10.46 ± 0.07

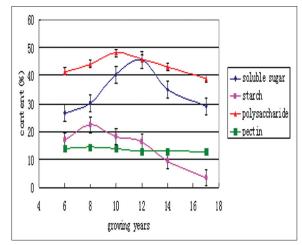


Figure 5: The accumulation trend of carbohydrate of LXSS

during the eighth year of growth then fell off. While the polysaccharide accumulated continually from the sixth to the tenth year, and reached a peak at the tenth year of growth, then fell off to the 17th year of growth. While the pectin accumulated continually from the sixth to the eighth year, and reached a peak at the eighth year of growth, then fell off a little and showed a tendency toward stabilization in the following years of growth. After SPSS analysis, we found that the content of starch and pectin and growing years were negatively correlated with the growing years with R values of -0.872 (P < 0.05) and -0.872 (P < 0.05). While the content of soluble sugar did not show significant correlation with growing years (P > 0.05), so did the total polysaccharide.

(2) The contents of ginsenosides and pectin are decreased a little with the growing year increasing and the weight of LXSS are increased with the growing year increasing [Table 3] either. Therefore, the total ginsenosides and pectin

Table 3: Total weight (g) of LXSS with different growing years ($n = 5$)						
Growing years	Total weight (g)					
6	3.00 ± 1.22					
8	2.62 ± 0.71					
10	3.85 ± 0.59					
12	5.52 ± 2.08					
14	4.87 ± 0.79					
17	6.31 ± 0.61					

of the same ginseng kept steady or increased a little during the period of growth.

Compared with the increasing of LXSS's weight, its content of starch fell off sharply. This maybe the reason for the tenacity of adventitious root of LXSS is similar to that of wild ginseng. As the cultivation environment and appearance of LXSS are similar to wild ginseng, the price of LXSS is higher than that of garden ginseng. That is in accordance with the application customs of TCM.

Correlations between ginsenosides, starch, and pectin in LXSS

The contents of total ginsenosides were positively correlated to the starch with R values of 0.889 (P < 0.05). Whereas the content of total ginsenosides were positively correlated to the pectin with R values of 0.967 (P < 0.01). The content of starch was positively correlated to the pectin with R values of 0.906 (P < 0.05). We know that ginsenosides that consist of aglycone and sugar are the main secondary metabolites of ginseng and play an important role in its life cycle. From these results, we could conjecture that sugars may mainly derive from the process of partial enzymatic hydrolysis of starch

Table 4: Significance difference test of the difference of component between LXSS and garden ginseng $(X \pm S)$

Component	Content (mg/g)		P	Component	Con	Content (%)	
	LXSS	Garden ginseng	arden ginseng		LXSS Garden ginsen		
Total ginsenoside*	24.06 ± 3.03	15.08 ± 2.83	0.000	Soluble sugar	34.42 ± 7.32	26.00 ± 17.76	0.254
PPT*	8.36 ± 0.81	5.16 ± 0.91	0.000	Starch*	14.38 ± 6.80	32.78 ± 8.79	0.001
PPD*	15.69 ± 2.22	9.92 ± 2.24	0.000	Polysaccharide	43.61 ± 3.25	51.22 ± 13.70	0.211
PPT/PPD	0.54 ± 0.03	0.53 ± 0.10	0.911	Pectin*	13.48 ± 0.65	10.61 ± 1.99	0.005
Rg₁/Re*	0.96 ± 0.10	0.73 ± 0.09	0.001				

^{*}Correlation is significant at the o.o1 level (two-tailed).

and pectin, which may explain the high correlation between total ginsenosides, starch, and pectin.

Correlations between ginsenosides, starch, and pectin in garden ginseng

The contents of total ginsenosides were positively correlated to the starch with R values of 0.608 (P>0.05). Whereas the contents of total ginsenosides were positively correlated to the pectin with R values of 0.771 (P<0.05). The contents of starch were positively correlated to the pectin with R values of 0.873 (P<0.01).

Garden ginseng's R values between total ginsenosides, starch, and pectin are lower than in LXSS, which may be relevant to manual intervention in the process of cultivation of garden ginseng.

Comparison of contents of ginsenosides and carbohydrate between LXSS and garden ginseng

The results [Table 4] showed statistically normal distribution and were analyzed by the t-test. We found that though there was no significant difference among soluble sugar, total polysaccharide, and the ratio of PPT and PPD in both LXSS and garden ginseng (P > 0.05), total ginsenosides, PPT, PPD, the ratio of Rg₁ and Re, and pectin of LXSS were significantly higher than that in garden ginseng (P < 0.01). Whereas starch of LXSS was significantly lower than in Garden ginseng (P < 0.01). The growing environment between LXSS and garden ginseng has shown a large difference and in comparison with garden ginseng, fertilizer and other manual interventions are not applied in LXSS. Maybe all about these can explain the difference mentioned above between LXSS and garden ginseng.

CONCLUSION

The results indicated that the contents of total ginsenosides, PPD, PPT, Rg₁, Re, Rb₁, Rc, Rb₂, Rd, starch, and pectin were negatively correlated with the growing years, whereas the contents of starch were positively correlated with the pectin. The total ginsenosides were positively correlated with starch and pectin, respectively; however, this cannot be

found in garden ginseng, maybe partly because of fertilizer and other manual intervention in process of cultivation of garden ginseng. The accumulation of ginsenosides and carbohydrate, especially starch and pectin varied from garden ginseng and LXSS. This research may provide the scientific basis for germplasm evaluation, the cultivation, and utilization of ginseng cultivated under mountainous forest.

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