

Separation and determination of chemical constituents in the volatile oil of three traditional Chinese crude drugs¹

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Abstract

The parts of Jilin ginseng growing on the ground, *Radix aucklandiae* and the citrus tangerina peels were extracted with ether, and yellow volatile oil 0.31 wt.%, 2.1 wt.% and 1.6 wt.% were obtained respectively. Their chemical constituents were separated and determined by gas chromatography and interfaced with mass spectrometry in an electron impact mode. © 1998 Elsevier Science B.V. All rights reserved.

Keywords: Jilin ginseng growing on the ground; *Radix aucklandiae*; Citrus tangerina; Volatile oil

1. Introduction

Ginseng is a product of the Araliaceae plant (*panax ginseng* C.A. Meyer). It is a traditional rare medical herb in China and the wild species of the herb found in the Jilin province, called Jilin ginseng, is the best and most famous in China as well as in foreign countries.

Since Garriques first isolated crude saponin from American ginseng many studies have been carried out on the ginseng [1–3]. The investigation and application of Jilin ginseng for the most part has concentrated on the ginseng saponin. The study on the volatile oil of ginseng

(GVO) did not begin until about 20 years ago [4,5]. In this paper, the parts of the Jilin ginseng growing on the ground (stems, leaves and flower buds) were extracted, and a yellow ginseng volatile oil was obtained. The GVO raw medicine was made, and its pharmacologic effect was examined. The results showed that it inhibited the growth of SGS-803 and S-180 cancer cells [6,7]. It was discovered that all the GVO constituents can be divided into several groups by thin layer chromatography according to their R_f values. The results show that one of the groups contains sesquiterpene compounds, which sends forth a delicate fragrance of ginseng. The sesquiterpenes make up the characteristic constituents of GVO. The sesquiterpene group in GVO was separated and determined by TLC and GC/MS methods.

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The *Radix Aucklandiae* is the drying rhizome of the compositae *Aucklandia Lappa Decne*. It is a perennial herb. It has a mild and bitter taste and has an analgesic effect on the stomach. A study on the volatile oil of the *Radix Aucklandiae* (AVO) has been reported. A study on the volatile oil from the Liaoning province in China has not been carried out up to now. In this paper, 22 kinds chemical constituents were extracted and determined by gas chromatography and mass spectrometry.

There are >1000 kinds of orange peels in China. Until now a study on the volatile oil (CVO) chemical constituents of the citrus tangerina peels from the Hunan province in China was not reported, as these orange peels were previously regarded as by-products and waste products. In this paper 20 chemical constituents have been obtained by gas chromatography and mass spectrometry. These include, α -pinene, β -pinene, carene, γ -terpinene, β -phelanthrene, sabinene, limonene, β -ocimene, Nerol, citral, α -multijugenol, β -elemene, cadinene, caryophyllene, α -humulene and nerolidol which are different from the constituents of the volatile oil from other provinces in China.

2. Experimental

2.1. Extraction of GVO, AVO and CVO

The parts of ginseng which grow on the ground from the Jilin province in China were treated by steam distillation, the distillate was collected and was extracted with ether, and a yellow oil was obtained. The content of GVO in the ginseng was 0.31%.

The *Radix Aucklandiae* from the Liaoning province in China was treated by steam distillation, the distillate was collected and was extracted with ether, and a yellow oil was obtained. The content of AVO in the *Radix Aucklandiae* was 2.1%.

The citrus tangerina peels from the Hunan province in China were treated by steam distillation, the distillate was collected and was extracted with ether, and a yellow oil was obtained. The content of CVO in the citrus tangerina was 1.6%.

2.2. Isolation of sesquiterpenes in GVO

The silica gel GF₂₅₄ thin layer chromatograph was used and hexane–ethylacetate (14:1) was the flowing phase. The silica gel G layers were impregnated with 5% A_gNO₃ and hexane–ethylacetate (28:1) was the mobile phase.

2.3. GC/MS analytical instruments and conditions of GVO

Hewlett Packard model 5890 gas chromatograph with a HP–5 capillary column with 25 m \times 0.25 mm \times 0.33 μ m film thickness was used. The flow rate of helium carrier gas was 1 ml min⁻¹. The temperature of gasification was 270°C. The column temperature programmed rate was 50–140°C (held for 15 min) at a rate of 10°C min⁻¹ and 260°C at a rate of 15°C min⁻¹ and then held. The sample splitting rate was 1:30.

Hewlett Packard model 5988A mass spectrometer was used. The ion source temperature was 200°C. The ion source was EI with an ionisation energy of 70 eV. The emission current was 300 μ A. Data was collected in full scan mode from m/z 40–600 in 1 s cycles.

2.4. GC/MS analytical instruments and conditions of AVO

Hewlett Packard model 5890 gas chromatograph with an OV–101 column with 25 m \times 0.25 mm \times 0.33 μ m film thickness was used. The flow rate of helium carrier gas was 2 ml min⁻¹. The temperature of gasification was 230°C. The column temperature programmed rate was 70–190°C at a rate of 4°C min⁻¹. The sample splitting rate was 1:80. Hewlett Packard model 5988A mass spectrometer was used. The ion source temperature was 190°C. The ion source was EI with an ionisation energy of 70 eV. The emission current was 300 μ A. Data was collected in full scan mode from 40–500 amu in 1 s cycles.

2.5. GC/MS analytical instruments and conditions of CVO

Hewlett Packard model 5890 gas chro-

Table 1
The chemical constituents of GVO, AVO and CVO

No.	Compound	Formula	Content (%)		
			GVO	AVO	CVO
1	Acetic acid ethyl ester	C ₄ H ₈ O ₂	a	4.35	a
2	Exthoxy butane	C ₆ H ₁₄ O	a	2.97	a
3	α -Terpinene	C ₁₀ H ₁₆	a	1.43	6.19
4	Carene	C ₁₀ H ₁₆	a	1.92	8.28
5	Camphene	C ₁₀ H ₁₆	a	0.34	a
6	α -Pinene	C ₁₀ H ₁₆	a	a	0.85
7	β -Pinene	C ₁₀ H ₁₆	a	a	2.65
8	γ -Terpinene	C ₁₀ H ₁₆	a	a	2.82
9	β -Phellandrene	C ₁₀ H ₁₆	a	a	2.64
10	Sabinene	C ₁₀ H ₁₆	a	a	2.35
11	α -Terpinene	C ₁₀ H ₁₆	a	a	6.19
12	β -Ocimene	C ₁₀ H ₁₆	a	a	1.91
13	Myrcene	C ₁₀ H ₁₆	a	a	0.93
14	Limonene	C ₁₀ H ₁₆ O	a	a	20.16
15	Citral	C ₁₀ H ₁₆ O	a	a	7.12
16	Nerol	C ₁₀ H ₁₈ O	a	a	5.16
17	Menthol	C ₁₀ H ₂₀ O	a	a	2.45
18	Eugenol	C ₁₀ H ₁₂ O ₂	a	a	1.18
19	Dihydro-actinidiolide	C ₁₁ H ₁₆ O ₂	a	12.35	a
20	α -Calacorene	C ₁₅ H ₂₀	a	12.15	a
21	β -Elemene	C ₁₅ H ₂₄	3.27	1.35	10.16
22	α -Humulene	C ₁₅ H ₂₄	0.70	0.36	1.16
23	α -Cubebene	C ₁₅ H ₂₄	0.29	a	a
24	α -Copaene	C ₁₅ H ₂₄	0.42	a	a
25	Aromadendrene	C ₁₅ H ₂₄	0.18	0.71	a
26	β -Gurjunene	C ₁₅ H ₂₄	1.25	a	a
27	α -Bisabolene	C ₁₅ H ₂₄	0.13	a	a
28	Cadinene	C ₁₅ H ₂₄	0.51	0.46	0.91
29	Eremophilene	C ₁₅ H ₂₄	1.28	a	a
30	β -Bourbonene	C ₁₅ H ₂₄	0.93	a	a
31	δ -Guaiene	C ₁₅ H ₂₄	a	2.75	0.84
32	Caryophyllene	C ₁₅ H ₂₄	10.84	a	0.75
33	Patchoulene	C ₁₅ H ₂₄	2.09	a	a
34	Farnesene	C ₁₅ H ₂₄	21.2	a	a
35	α -Santalene	C ₁₅ H ₂₄	5.57	a	a
36	α -Bergamotene	C ₁₅ H ₂₄	0.26	a	a
37	β -Santslene	C ₁₅ H ₂₄	0.86	a	a
38	β -Maallene	C ₁₅ H ₂₄	0.79	11.41	a
39	β -Cubeber	C ₁₅ H ₂₄	0.25	a	a
40	Alloaromadendrene	C ₁₅ H ₂₄	0.77	a	a
41	α -Guaiene	C ₁₅ H ₂₄	0.26	a	a
42	β -Bisabolene	C ₁₅ H ₂₄	1.47	a	a
43	α -Gurjunene	C ₁₅ H ₂₄	0.32	a	a
44	Isolongifolene	C ₁₅ H ₂₄	0.86	a	a
45	Selinene	C ₁₅ H ₂₄	0.69	0.60	a
46	β -Cedrene	C ₁₅ H ₂₄	a	1.08	a
47	β -Curcumene	C ₁₅ H ₂₄	a	1.15	a
48	β -Himachalene	C ₁₅ H ₂₄	a	1.28	a
49	α -Costol	C ₁₅ H ₂₄ O	a	6.21	a
50	α -Multijungenol	C ₁₅ H ₂₄ O	a	a	1.14
51	Nerolidol	C ₁₅ H ₂₄ O	a	a	0.81
52	Dehydro-costuslactone	C ₁₅ H ₁₈ O	a	25.68	a

^a The compound is not present in the volatile oil.

matograph with an OV-101 column with $25\text{ m} \times 0.25\text{ mm} \times 0.33\text{ }\mu\text{m}$ film thickness was used. The flow rate of helium carrier gas was 1 ml min^{-1} . The temperature of gasification was 250°C . The column temperature programmed rate was $70\text{--}220^\circ\text{C}$ at a rate of 5°C min^{-1} . The sample splitting rate was 1:60.

Hewlett Packard model 5988A mass spectrometer was used. The ion source temperature was 220°C . The ion source was EI with an ionisation energy of 70 eV. The emission current was $300\text{ }\mu\text{A}$. Data was collected in full scan model from 40–600 amu in 1 s cycles.

3. Results

Analytical results showed that there was a group of 24 sesquiterpene compounds in GVO of the parts of the ginseng growing on the ground with formula $\text{C}_{15}\text{H}_{24}$ and m/z 204 (Table 1). The weight of the 24 identified sesquiterpene constituents was 47.14% of the GVO.

The percentage of every sesquiterpene in GVO has been calculated as a % by using the integral programme. The ginseng sesquiterpenes were identified according to checking up by means of a computer with the Wiley library, contrasting with EPA and spice mass spectrogram, and elucidating the spectrogram from the principles of MS [8–10].

Nineteen chemical constituents have been obtained by analysing the *Radix Aucklandiae* volatile oil from the Liaoning province in China. Nineteen chemical compound constituents of AVO are listed in Table 1. The weight of the 19 identified constituents was 77.54% of the AVO.

Twenty two chemical constituents of CVO have been obtained using the above methods. The weight of the 22 identified constituents was 86.75% of the CVO.

4. Discussion

Ginseng, *Radix Aucklandiae* and Citrus Tangerina are all natural traditional Chinese crude drugs. The parts of the ginseng growing on the ground, that is the stems, leaves and flower buds of ginseng were the by-product in a ginseng garden. The peels of citrus Tangerina were also the by-product of citrus. The experimental results show that the chemical constituents of GVO and CVO belong to the terpinene compounds. The volatile oils can be utilised for medicinal purposes and as foodstuff.

We see in Table 1 that the volatile oil of all three traditional Chinese crude drugs is a mixture. GVO contains 24 compounds, AVO contains 19 compounds and CVO contains 22 compounds, most of the compounds belong to the terpinenes, a few the compounds belong to the aromatic and aliphatic compounds. We noticed that some compounds are the same among the GVO, AVO and CVO, for example, β -elemene, α -humulene and cadinene.

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