Preparative Separation of Flavanones and Terpenoids from Olibanum by High-Speed Counter-Current Chromatography

H. W. Zhao^{1†}, Y. L. Geng¹, H. Zhu¹, P. Yang² and J. Q. Yu^{1*}

¹Shandong Key Laboratory of TCM Quality Control Technology, Shandong Analysis and Test Center, Jinan, 250014, P. R. China

²Senkang Sanfeng Biological Engineering Technology Co. Ltd., Jinan, 250014, P. R. China

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Seven compounds, including two flavanones, dihydrokaempferol (**1**) and naringenin (**2**), and five terpenoids, boscartol A (**3**), 3,7-dioxo-tirucalla-8,24-dien-21-oic acid (**4**), 3 α -acetoxyl-7-oxo-tirucalla-8,24-dien-21-oic acid (**5**), 11-keto- β -boswellic acid (**6**), and acetyl-11-keto-boswellic acid (**7**), have been purified by high-speed counter-current chromatography (HSCCC) from olibanum. For the separation, from 250 mg of the crude extract, 3.1 mg of **1** (95.2% purity), 2.7 mg of **2** (96.1% purity), 9.1 mg of **3** (96.7% purity), 4.5 mg of **4** (95.3% purity), 5.4 mg of **5** (96.3% purity), 48.1 mg of **6** (96.8% purity), and 45.5 mg of **7** (98.1% purity) were obtained by HSCCC with petroleum ether–ethyl acetate–methanol–water (1:0.8:1.1:0.6, v/v). The structures of these seven compounds were elucidated by a combination of electrospray ionization mass spectrometry (ESI–MS) and extensive nuclear magnetic resonance (NMR) spectroscopic.

Keywords: Olibanum, flavanone, terpenoid, separation, high-speed counter-current chromatography

Introduction

Olibanum, the exuded gum resin of *Boswellia carterii* Birdw., well known as a folk crude drug in traditional Chinese medicine (TCM), has been used to treat chest stuffiness and pains, dysmenorrhea, rheumatic arthralgia, and amenorrhea, as well as traumatic injuries [1]. Previous investigations on chemical and pharmacological studies of the resin have led to the isolation and identification of a number of terpenoids, including triterpenes, triterpene acids, diterpenes, and monoterpenoids, etc., with the first three kinds having been suggested to be the major active constituents, which have been revealed to possess cytotoxic, analgesic, anti-inflammatory, antifungal, antibacterial, and antidiabetic activities [2, 3]. However, terpenoids and other compounds from olibanum had been isolated mostly based on some conventional techniques, such as silica gel column chromatography separation due to their lower polarities [4], which are waste organic solvent, tedious, time consuming, and often result in irreversible absorption of compounds [5, 6]. Thus, effective separation methods for components from olibanum are of great importance. As far as we know, no reports have been published on the separation of flavanones and terpenoids by high-speed counter-current chromatography (HSCCC) from olibanum.

HSCCC is a liquid–liquid partition chromatography, which has now been widely applied in the separation of various compounds [7–11]. Generally speaking, HSCCC allows no solid support and separation of compounds between the stationary and mobile phases constituted by two immiscible solvent systems, as well as possesses high loading capacity, low economic cost, and no irreversible absorption of samples. Herein, this paper describes the successful isolation of seven compounds, including two flavanones, dihydrokaempferol (**1**) and naringenin (**2**), and five terpenoids, boscartol A (**3**), 3,7-dioxo-tirucalla-8,24-dien-21-oic acid (**4**), 3 α -acetoxyl-7-oxo-tirucalla-8,24-dien-21-oic acid (**5**), 11-keto- β -boswellic acid (**6**), and acetyl-11-keto-boswellic acid (**7**) (Figure 1) from olibanum by HSCCC in a single run.

Experimental

Reagents and materials. Acetonitrile of HPLC grade used in HPLC analysis was purchased from Fisher Scientific (Fair Lawn, NJ, USA), and other solvents, including methanol, alcohol, ethyl acetate, and petroleum ether were of analytical grade (Fuyu Fine Chemical Co., Ltd, Tianjin, China). The water used was deionized by an osmosis Milli-Q system (Millipore, Bedford, MA, USA).

Olibanum, the gum resin of *B. carterii* Birdw. originating from Ethiopia, was purchased from Bozhou Medicine market in Anhui province and authenticated by Prof. Fengqin Zhou (Shandong University of Traditional Chinese Medicine). The voucher (No. RX201412) was stored in Shandong Key Laboratory of TCM Quality Control Technology, Shandong Analysis and Test Center, Jinan, Shandong, China.

Apparatus. The HSCCC separation was conducted on a TBE-300A high-speed counter-current chromatograph (Tauto Biotechnology, Shanghai, China), which was equipped with a 300 mL polytetrafluoroethylene (PTFE) multilayer coil (diameter of the PTFE tube as 1.6 mm) as well as a 20 mL manual sample loop. The rotation speed of the column coil could be adjustable from 0 to 1000 rpm. The HSCCC apparatus was also equipped with four other instrument modules, including a TBP-5002 constant-flow pump (Tauto Biotechnology, Shanghai, China), a 8823A-UV Monitor at 254 nm (Beijing Emilion Technology, Beijing, China), a Model 3057 portable recorder (Yokogawa, Sichuan Instrument Factory, Sichuan, China), and a DC-0506 low constant temperature bath (Tauto Biotechnology, Shanghai, China) to maintain the temperature at 25 °C.

A Waters e2695 equipment with a Waters 2695 quaternary-solvent delivery system, a Waters 2998 photodiode array detector (PAD), an automatic sample injection, a Waters 2695 column oven, and an Empower 3 ChemStation was used to analyze the crude extract and the collected fractions. The column used was a Waters WAT054275-C₁₈ column (250 mm $\times$ 4.6 mm, i.d. 5 μ m, USA). Nuclear magnetic resonance (NMR) spectra were performed on a Bruker AV-400 spectrometer (Bruker BioSpin, Rheinstetten, Germany), and electrospray ionization mass spectrometry (ESI–MS) experiments were performed on an Agilent 6520 Q-TOF (Agilent, USA).

* Author for correspondence: yujinqian87528@126.com

† Current Affiliation: College of Pharmacy, Shandong University of Traditional Chinese Medicine, Jinan, 250014, P. R. China.

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Figure 1. Chemical structures of compounds 1–7.

Preparation of crude sample. The olibanum was powdered and sieved (40–60 mesh) and then weighed exactly (100 g) in a 500 mL flask; then, 200 mL of ethyl acetate was added. The extraction procedure was carried out for three times (1 h, 0.5 h, 0.5 h) at 60 °C. Finally, the extract was concentrated under reduced pressure to provide 10.2 g of crude sample, storing at 2–8 °C for the subsequent CCC separation. The HPLC chromatogram of the crude ethyl acetate extract is presented in Figure 2.

HSCCC separation procedure. For HSCCC experiment, the separation column was initiated by filling with the upper phase at 20.0 mL/min, and then the column was rotated at 800 rpm, while the mobile phase was pumped into the column at 2.0 mL/min in the head to tail direction. After the equilibration reached, the sample solution (each 5 mL for upper and lower phase) containing 250 mg of sample was injected. Fractions collected manually every 5 min were monitored online at 254 nm with an ultraviolet (UV) detector.

Figure 2. HPLC chromatograms of the extraction sample from olibanum with different UV detection wavelength (a: 248 nm; b: 287 nm). HPLC conditions: column: Waters WAT054275- C_{18} column (250 mm $\times$ 4.6 mm, i.d. 5 μ m); mobile phase: water (A) and acetonitrile (B) (0–20 min, 40%–60% B; 20–30 min, 60%–80% B; 30–40 min, 80%–100% B; 40–50 min, 100% B); column temperature: 25 °C; flow rate: 1.0 mL/min; injection volume: 10 μ L; peak 1: dihydrokaempferol (1), peak 2: naringenin (2), peak 3: boscartol A (3), peak 4: 3,7-dioxo-tirucalla-8,24-dien-21-oic acid (4), peak 5: 3 α -acetoxyl-7-oxo-tirucalla-8,24-dien-21-oic acid (5), peak 6: 11-keto- β -boswellic acid (6), and peak 7: acetyl-11-keto-boswellic acid (7).

After the compounds were eluted out, the solvent in the column was continuously forced out by nitrogen, which was collected for computing the retention of the stationary phase and subsequent HPLC analysis.

HPLC analysis and identification of CCC fractions. The crude sample and each peak fraction from the separation procedures were analyzed by HPLC with a Waters WAT054275- C_{18} column (250 mm $\times$ 4.6 mm i.d., 5 μ m) at 248 and 287 nm, respectively, on a Waters e2695 equipment. The mobile phase was water (A) and acetonitrile (B), and the gradient elution mode was as follows: 0–20 min, 40%–60% B; 20–30 min, 60%–80% B; 30–40 min, 80%–100% B; 40–50 min, 100% B with a flow rate of 1.0 mL/min. The chemical structures of these compounds were elucidated by extensive spectroscopic measurements and comparison with data reported in literatures.

Results and Discussion

Optimization of the HSCCC conditions. The separation of the crude extract from olibanum was carried out by HSCCC. To obtain a successful separation, choosing the satisfied stationary phase and mobile phase was the most important step, which should display suitable K_D values (usually expected to be between 0.5 and 2.0) [12]. According to the structure characteristics of flavanones and terpenoids, three solvent systems of petroleum ether–ethyl acetate–methanol–water (1:0.8:1.1:0.6, 1:0.6:1.1:0.6, and 1:0.4:1.1:0.6, v/v) was optimized with the ratio of ethyl acetate varying from 0.4 to 0.8 on the basis of the previous research [13–15]. The K_D values and the separation factors of the target compounds in the three solvent systems were detected and calculated (shown in Table 1). First, when selecting the solvent system of petroleum ether–ethyl acetate–methanol–water (1:0.4:1.1:0.6, v/v), it was found that compounds 1–6 were mainly distributed in the lower layer, which meant that the polarity of the system was much weaker than the polarity of the sample. Therefore, the ratio of ethyl acetate was increased to increase the polarity of the system. Eventually, when the ratio of ethyl acetate increased up to 0.8 for the above solvent system, the desired one was obtained, giving suitable K_D values and separation factors. Though the separation factor between peaks 2 and 3 was actually greater at the ratio of 1:0.4:1.1:0.6 rather than the system of 1:0.8:1.1:0.6, and the separation factor between peaks 3 and 4 was actually greater at the ratio of 1:0.6:1.1:0.6 rather than the system of 1:0.8:1.1:0.6, it was found that only the 0.8 ratio of ethyl acetate could produce successfully separation of these seven compounds.

As shown in Figure 3, the separation time between compounds 3 and 6 was more than an hour, which was considered as a waste. To settle this problem, gradient elution mode was chosen to speed the isolation process. The solvent system of petroleum ether–ethyl acetate–methanol–water with volume ratio of 1:0.8:1.3:0.6 was chosen as the gradient solvent to separate compounds 5–7, in which the ratio of methanol increased up to 1.3 to speed the separation. Finally, petroleum ether–ethyl acetate–methanol–water with volume ratio of 1:0.8:1.1:0.6 and 1:0.8:1.3:0.6 were assigned for HSCCC separation with a

Figure 3. HSCCC chromatogram for the separation of the crude extract. Experimental conditions: solvent system: petroleum ether–ethyl acetate–methanol–water (1:0.8:1.1:0.6, v/v); the upper organic phase as the stationary phase; revolution speed: 800 rpm; flow rate: 2.0 mL/min; sample size: 250 mg; UV detection wavelength: 254 nm; retention of stationary phase: 67.7%.

stepwise mode. A series of experiments about system changing time was investigated. Although the mobile phase was replaced after 3.0 hr, compound 5 could not be well separated from compound 6 (Figure 4). From the above, the HSCCC separation with one solvent system was found to be the best for the separation of the seven compounds.

Besides the solvent system and elution mode, two other factors, the revolution speed of the separation column and the flow rate of the mobile phase, were also investigated. Different revolution speed (700, 800, and 900 rpm) and different flow rates (2.0, 5.0, and 10.0 mL/min) were examined. The revolution speed could affect the retention of stationary phase, and the lower speed will lead to a lower stationary phase retention. However, when a high revolution speed was used, emulsification could easily appear. The results revealed that, when the revolution speed was 800 rpm, a better retention of stationary phase of 66.7% could be achieved. When the flow rate of 2.0 mL/min was tested, all the seven compounds could be well separated within 7 h. When a large flow rate of 5.0 mL/min was used, only compounds 1, 3, 6, and 7 could be obtained within 4 h, as well as a larger flow rate of 10.0 mL/min could only obtain compounds 6 and 7 within 3 h. The results revealed that, when the flow rate was 2.0 mL/min, satisfactory separation efficiency and peak resolutions could be achieved.

Separation of the crude extract of terpenoids by HSCCC. As shown in Figure 3, each peak of the target compound was

Figure 4. HSCCC separation of the crude extract using a stepwise mode. Experimental conditions: solvent system: first, petroleum ether–ethyl acetate–methanol–water (1:0.8:1.1:0.6, v/v), the upper organic phase as the stationary phase; at 3.1 h, replaced the mobile phase by petroleum ether–ethyl acetate–methanol–water (1:0.8:1.3:0.6, v/v) lower phase; revolution speed: 800 rpm; flow rate: 2.0 mL/min; sample size: 250 mg; UV detection wavelength: 254 nm; retention of stationary phase: 55.7%.

Table 1. Partition coefficients (K_D) and separation factors (α) of the target compounds in different solvent systems

Solvent system Pet–EtAc–MeOH–H ₂ O	Peak no.													
	1 (K_D)	α	2 (K_D)	α	3 (K_D)	α	4 (K_D)	α	6 (K_D)	α	5 (K_D)	α	7 (K_D)	α
1:0.4:1.1:0.6	0.09	1.44	0.13	1.46	0.19	1.10	0.21	1.42	0.30	2.00	0.60	6.4	3.84	
1:0.6:1.1:0.6	0.14	1.42	0.20	1.10	0.22	1.50	0.33	1.48	0.49	1.59	0.78	6.12	4.78	
1:0.8:1.1:0.6	0.28	1.57	0.44	1.25	0.55	0.95	0.52	1.40	0.73	1.71	1.25	4.22	5.27	
1:0.8:1.3:0.6	–	–	–	–	–	–	–	–	0.54	1.56	0.84	5.03	4.23	

separated with good resolution in the typical chromatogram of separation of the crude sample by HSCCC. Because compound **7** had the lowest polarity among all the compounds, as well as a largest K_D value of 5.27, it will take a lengthy time to separate **7**. To reduce the separation time, after the first six compounds were removed, compound **7** was eluted by forcing out of the stationary phase by nitrogen. Each peak fraction was analyzed by HPLC. As a result, 3.1 mg of **1** (95.2% purity at 287 nm) (collected from 70 to 75 min), 2.7 mg of **2** (96.1% purity at 287 nm) (collected from 80 to 90 min), 9.1 mg of **3** (96.7% purity at 248 nm) (collected from 165 to 180 min), 4.5 mg of **4** (95.3% purity at 248 nm) (collected from 140 to 150 min), 5.4 mg of **5** (96.3% purity at 248 nm) (collected from 330 to 340 min), 48.1 mg of **6** (96.8% purity at 248 nm) (collected from 275 to 310 min), and 45.5 mg of **7** (98.1% purity at 248 nm) (collected from 315 to 340 min) could be obtained from 250 mg crude extract.

Structure identification of the isolated compounds. The chemical structures of the isolated flavanones and terpenoids were identified by comparison of their spectroscopic data with the reported values, including ESI-MS, $^1\text{H-NMR}$, and $^{13}\text{C-NMR}$ data. As a result, seven compounds were elucidated, including two flavanones, dihydrokaempferol (**1**) and naringenin (**2**), and five terpenoids, boscactol A (**3**), 3,7-dioxo-tirucalla-8,24-dien-21-oic acid (**4**), 3 α -acetoxyl-7-oxo-tirucalla-8,24-dien-21-oic acid (**5**), 11-keto- β -boswellic acid (**6**), and acetyl-11-keto-boswellic acid (**7**) [1, 16–18]. The two flavanones were obtained as yellow amorphous powder, and the five terpenoids were all obtained as white amorphous powder.

Dihydrokaempferol (1): ESI-MS, m/z 311.3 $[\text{M} + \text{Na}]^+$. $^1\text{H-NMR}$ (DMSO- d_6 , 400 MHz) δ : 4.58 (1H, d, $J = 11.2$ Hz, H-3), 5.04 (1H, d, $J = 11.2$ Hz, H-2), 5.86 (1H, s, H-6), 5.91 (1H, s, H-8), 6.78 (2H, d, $J = 8.4$ Hz, H-3', 5'), 7.30 (2H, d, $J = 8.4$ Hz, H-2', 6'). $^{13}\text{C-NMR}$ (DMSO- d_6 , 100 MHz) δ : 70.4 (C-3), 81.8 (C-2), 94.4 (C-8), 95.3 (C-6), 100.2 (C-10), 114.1 (C-3', 5'), 127.3 (C-1'), 128.5 (C-2', 6'), 157.2 (C-4'), 163.3 (C-9), 165.5 (C-5), 167.7 (C-7), 197.0 (C-4).

Naringenin (2): ESI-MS, m/z 273.3 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (DMSO- d_6 , 400 MHz) δ : 2.78 (1H, dd, $J = 3.0, 16.8$ Hz, H_b-3), 3.27 (1H, dd, $J = 12.8, 16.8$ Hz, H_a-3), 5.44 (1H, dd, $J = 3.0, 12.8$ Hz, H-2), 5.88 (2H, s, H-6, 8), 6.79 (2H, d, $J = 8.4$ Hz, H-3', 5'), 7.32 (2H, d, $J = 8.4$ Hz, H-2', 6'). $^{13}\text{C-NMR}$ (DMSO- d_6 , 100 MHz) δ : 42.5 (C-3), 78.9 (C-2), 95.4 (C-8), 96.3 (C-6), 102.2 (C-10), 115.6 (C-3', 5'), 128.8 (C-2', 6'), 129.3 (C-1'), 158.2 (C-4'), 163.4 (C-9), 164.0 (C-5), 167.2 (C-7), 196.8 (C-4).

Boscactol A (3): ESI-MS, m/z 325.3 $[\text{M} + \text{Na}]^+$. $^1\text{H-NMR}$ (DMSO- d_6 , 400 MHz) δ : 1.09 (3H, s, Me-14), 1.11 (3H, s, Me-11), 1.73 (3H, s, Me-20), 4.01 (2H, s, H-19), 4.63 (1H, s, H_b-12), 4.65 (1H, s, H_a-12), 5.24 (1H, d, $J = 15.2$ Hz, H-15), 5.75 (1H, d, $J = 11.2$ Hz, H-17), 6.21 (1H, dd, $J = 11.2, 15.2$ Hz, H-16). $^{13}\text{C-NMR}$ (DMSO- d_6 , 100 MHz) δ : 13.1 (C-14), 21.8 (C-20), 24.3 (C-13), 26.4 (C-8), 26.5 (C-11), 26.9 (C-2), 28.7 (C-7), 31.8 (C-6), 38.6 (C-9), 41.8 (C-3), 53.1 (C-1), 53.2 (C-5), 60.2 (C-19), 79.2 (C-4), 106.8 (C-12), 120.4 (C-16), 126.9 (C-17), 135.2 (C-18), 143.7 (C-15), 153.4 (C-10).

3,7-Dioxo-tirucalla-8,24-dien-21-oic acid (4): ESI-MS, m/z 469.3 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (DMSO- d_6 , 400 MHz) δ : 0.70 (3H, s, Me-18), 0.89 (3H, s, Me-30), 0.97 (3H, s, Me-28), 1.03 (3H, s, Me-29), 1.21 (3H, s, Me-19), 1.53 (3H, s, Me-27), 1.64 (3H, s, Me-26), 5.08 (1H, t, $J = 6.8$ Hz, H-24). $^{13}\text{C-NMR}$ (DMSO- d_6 , 100 MHz) δ : 16.0 (C-18), 17.9 (C-27), 17.9 (C-19), 21.5 (C-29), 23.7 (C-11), 24.6 (C-26), 24.7 (C-23), 25.9 (C-28), 26.0 (C-16), 27.3 (C-12), 28.0 (C-15), 31.5 (C-22), 32.7 (C-30), 34.6 (C-2), 34.8 (C-1), 36.1 (C-6), 39.1 (C-10), 44.3 (C-13), 45.5 (C-17), 47.1 (C-4), 47.3 (C-14), 47.7 (C-20), 48.8 (C-5), 124.3 (C-24), 131.6 (C-25), 138.2 (C-8), 164.7 (C-9), 177.6 (C-21), 196.9 (C-7), 214.3 (C-3).

3 α -Acetoxyl-7-oxo-tirucalla-8,24-dien-21-oic acid (5): ESI-MS, m/z 497.4 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (DMSO- d_6 , 400 MHz) δ : 0.73 (3H, s, Me-18), 0.79 (3H, s, Me-30), 0.88 (3H, s, Me-28), 0.95 (3H, s, Me-29), 1.02 (3H, s, Me-19), 1.54 (3H, s, Me-27), 1.64 (3H, s, Me-26), 4.56 (1H, brs), 5.08 (1H, t, $J = 7.2$ Hz, H-24), 12.1 (1H, brs, COOH), 2.01 (3H, s, CH₃CO-). $^{13}\text{C-NMR}$ (DMSO- d_6 , 100 MHz) δ : 16.1 (C-18), 17.9 (C-27), 18.3 (C-19), 21.5 (C-11), 22.9 (C-2), 23.2 (C-26), 24.6 (C-30), 25.9 (C-23), 26.0 (C-28), 27.1 (C-29), 27.4 (C-16), 28.0 (C-12), 29.5 (C-15), 31.5 (C-22), 32.7 (C-1), 35.4 (C-6), 36.7 (C-4), 39.2 (C-10), 43.6 (C-13), 44.3 (C-14), 45.6 (C-20), 47.2 (C-17), 47.8 (C-5), 76.6 (C-3), 124.3 (C-24), 131.6 (C-25), 138.0 (C-8), 166.1 (C-9), 177.6 (C-21), 197.0 (C-7), 21.4 (CH₃CO-), 170.3 (CH₃CO-).

11-Keto- β -boswellic acid (6): ESI-MS, m/z 471.3 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (CDCl₃, 400 MHz) δ : 0.81 (3H, d, $J = 6.0$ Hz, Me-29), 0.84 (3H, s, Me-30), 0.96 (3H, s, Me-28), 1.15 (3H, s, Me-25), 1.20 (3H, s, Me-26), 1.33 (3H, s, Me-27), 1.36 (3H, s, Me-23), 4.10 (1H, brs, H-3), 5.57 (1H, brs, H-12). $^{13}\text{C-NMR}$ (CDCl₃, 100 MHz) δ : 13.2 (C-25), 17.4 (C-29), 18.4 (C-26), 18.8 (C-6), 20.5 (C-27), 21.1 (C-28), 24.3 (C-23), 26.2 (C-2), 27.2 (C-16), 27.5 (C-21), 28.9 (C-30), 30.9 (C-7), 32.9 (C-15), 34.0 (C-1), 37.5 (C-10), 39.3 (C-19), 39.3 (C-20), 40.9 (C-22), 43.8 (C-8), 45.1 (C-14), 47.3 (C-4), 48.8 (C-5), 59.0 (C-18), 60.4 (C-9), 70.5 (C-3), 130.5 (C-12), 165.1 (C-13), 182.3 (C-24), 199.6 (C-11).

Acetyl-11-keto-boswellic acid (7): ESI-MS, m/z 513.5 $[\text{M} + \text{H}]^+$. $^1\text{H-NMR}$ (CDCl₃, 400 MHz) δ : 0.79 (3H, d, $J = 6.0$ Hz, Me-29), 0.82 (3H, s, Me-30), 0.94 (3H, s, Me-28), 1.14 (3H, s, Me-25), 1.19 (3H, s, Me-26), 1.34 (3H, s, Me-27), 1.23 (3H, s, Me-23), 2.08 (3H, s, CH₃CO-), 5.30 (1H, brs, H-3), 5.55 (1H, brs, H-12). $^{13}\text{C-NMR}$ (CDCl₃, 100 MHz) δ : 13.2 (C-25), 17.4 (C-29), 18.4 (C-26), 18.8 (C-6), 20.5 (C-30), 21.1 (C-27), 23.5 (C-2), 23.8 (C-23), 27.2 (C-16), 27.4 (C-15), 28.8 (C-28), 32.8 (C-7), 34.6 (C-1), 37.4 (C-10), 39.3 (C-19), 39.3 (C-20), 40.9 (C-22), 43.8 (C-8), 45.1 (C-14), 46.5 (C-21), 46.5 (C-4), 50.4 (C-5), 59.0 (C-18), 60.3 (C-9), 73.1 (C-3), 130.5 (C-12), 166.1 (C-13), 181.8 (C-24), 199.4 (C-11), 21.3 (CH₃CO-), 170.3 (CH₃CO-).

Conclusions

In this paper, an efficient technique for the separation of flavanones and terpenoids from olibanum by HSCCC has been developed for the first time. At last, seven compounds, including two flavanones, naringenin (**1**) and dihydrokaempferol (**2**), and five terpenoids, boscactol A (**3**), 3,7-dioxo-tirucalla-8,24-dien-21-oic acid (**4**), 3 α -acetoxyl-7-oxo-tirucalla-8,24-dien-21-oic acid (**5**), 11-keto- β -boswellic acid (**6**), and acetyl-11-keto-boswellic acid (**7**) were successfully obtained by HSCCC in a single run. The purities of these compounds were all over 95.0%, which demonstrated that HSCCC as a powerful preparative chromatography is an efficient method for isolating components from olibanum.

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