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黄瓜藤的化学成分研究

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摘 要:从丽江产黄瓜藤甲醇提取物的氯仿部位分离得到 9个化合物,经理化和波谱分析鉴定为 α-菠甾醇(**1**)、α-菠甾醇-3-*O*-β-*D*-葡萄糖苷(**2**)、β-谷甾醇(β-sitosterol, **3**)、豆甾-7-烯-3-*O*-β-*D*-葡萄糖苷(**4**)、22-亚甲基-9, 19-环羊毛甾烷-3β-醇(**5**)、(2*S*, 3*S*, 4*R*, 10*E*)-2-(2′,3′-二羟基二十四烷酰氨基)-10-十八烯-1, 3, 4-三醇(**6**)、(2*S*, 3*S*, 4*R*, 10*E*)-2-[(2′*R*)-2-羟基二十四烷酰氨基]-10-十八烯-1, 3, 4-三醇(**7**)、(2*S*, 3*S*, 4*R*, 10*E*)-1-(β-*D*-葡萄糖苷)-2-[(2′*R*)-2-羟基二十四烷酰氨基]-10-十八烯-1, 3, 4-三醇(**8**)、大豆脑苷(**9**),除化合物 **3**外,其它化合物均为首次从该植物中分离得到。

关键词:黄瓜;藤;化学成分

中图分类号: R284.1; Q946.8; S642.2 **文献标识码**: A

Chemical Constituents from Stems of *Cucumis sativus* L.

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Abstract: Nine compounds were isolated from the chloroform fraction of the methanol extract of *Cucumis sativus* L. stems, and their structures were elucidated by physicochemical properties and spectroscopic analysis as α-spinasterol (**1**), α-spinasterol 3-*O*-β-*D*-glucopyranoside (**2**), β-sitosterol (**3**), stigmast-7-en-3-*O*-β-*D*-glucopyranoside (**4**), 22-methylene-9, 19-cyclolanostan-3β-ol (**5**), (2*S*, 3*S*, 4*R*, 10*E*)-2-(2′,3′-dihydroxytetracosanoylamino)-10-octadecene-1, 3, 4-triol (**6**), (2*S*, 3*S*, 4*R*, 10*E*)-2-[(2′*R*)-2-hydroxy tetracosanoylamino]-10-octadecene-1, 3, 4-triol (**7**), (2*S*, 3*S*, 4*R*, 10*E*)-1-(β-*D*-glucopyranosyl)-2-[(2′*R*)-2-hydroxytetracosanoylamino]-10-octadecene-1, 3, 4-triol (**8**), soya-cerebroside I (**9**). By literature study, except for compound **3**, the others were isolated from this plant for the first time.

Key words: *Cucumis sativus* L; stem; chemical constituent

Introduction

Cucumis sativus L, which belongs to Cucurbitaceae, is now widely planted in the temperate and tropical zones, including all the districts in China [1]. It is one of the most important vegetables, and the stems have been used in traditional Chinese medicine for its anti-inflammatory activity. According to the book "Ben Cao Gang Mu" edited by Shizhen Li of Ming Dynasty of ancient China, the stems can expand blood vessel and reduce blood pressure [2]. However, very little is known about its chemical constituents from the stems, though some reports suggested the presence of steroids and phenolics in this plant [3,4]. The present study involved isolation and identification of nine compounds from the chloroform fraction of the methanol extract from stems of *C. sativus*.

Experimental

General

Melting points were determined using an XRC-1 micro-melting point apparatus, and uncorrected. NMR spectra (^{1}H NMR, ^{13}C NMR and DEPT) were recorded on either Bruker AV-400 or Bruker DRX-500 NMR instruments. The chemical shifts were expressed in ppm as δ

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values relative to tetramethylsilane (TMS) with TMS as an internal standard. MS spectra were recorded on VG Auto Spec-3000 spectrometer. Column chromatography was performed on either silica gel (200-300 mesh) or Sephadex LH-20 (25-100 μm, Pharmacia Company). TLC was performed on pre-coated silica gel F_{254} plates (Qingdao Haiyang Chemical Company, China), and detection was provided by UV at 254 nm and spraying with 10% H_2SO_4-EtOH followed by heating at 100 °C.

Plant material

Stems of *Cucumis sativus* L. were collected in August, 2005 in Lijiang of Yunnan Province in the southwest of China, which were identified by Prof. Shukun Chen of Kunming Institute of Botany. The stems were left to dry in the shade at room temperature to a constant weight. A voucher specimen was deposited in the Herbarium of the Department of Taxonomy, Kunming Institute of Botany, Chinese Academy of Sciences.

Extraction and isolation

The dry stems (4.5 kg) of *C. sativus* were ground into powder and then extracted for three times with methanol under reflux at 60 °C, 4 h extraction for each time. The combined filtrate was concentrated *in vacuo* at 50 °C using a rotary evaporator to afford a residue as methanol crude extract (330 g), which was further suspended in water and extracted successively with chloroform, and then with *n*-butanol to give chloroform fraction (65 g) and *n*-butanol fraction (60 g). Chloroform fraction was then subjected to column chromatography on a silica gel (1200 g, 200-300 mesh), eluting with a gradient mixture of $CHCl_3$-MeOH (from 50 ∶1 to 0 ∶1, v/v) to yield five sub-fractions based on TLC analysis. Sub-fraction 1 (4.8 g) was further chromatographied on a silica gel (200-300 mesh) using petroleum ether-acetone (from 10 ∶1 to 2 ∶1, v/v) as an eluant to yield compounds **1** (5 mg) and **3** (202 mg). Compound **5** (41 mg) was isolated from sub-fraction **2** (4.6 g) by silica gel (200-300 mesh) column eluted with petroleum ether-acetone (5 ∶1, v/v). Sub-fraction 3 (3.5 g) was repeatedly chromatographyed over silica gel (200-300 mesh) eluted with $CHCl_3$-MeOH (from 30 ∶1 to 10 ∶1, v/v), and then on Sephadex LH-20 eluting with MeOH to yield compounds **6** (36 mg) and **7** (146 mg). Sub-fraction **4** (9.3 g) was chromatographyed on silica gel (200-300 mesh) using $CHCl_3$-MeOH (from 20 ∶1 to 5 ∶1, v/v) as an eluant to give the compounds **2** (886 mg) and **4** (35 mg). Compounds **8** (674 mg) and **9** (164 mg) were obtained from sub-fraction **5** (13 g) by repeated silica gel chromatography eluting with $CHCl_3$-MeOH (from 10 ∶1 to 2 ∶1, v/v) solvent system.

Results and Discussion

α-Spinasterol (1) White needles (MeOH), $C_{29}H_{48}O$, mp. 158-159 °C, ESI-MS *m/z*: 413 $[M+H]^+$. ^{1}H NMR ($CDCl_3$, 500 MHz) δ: 5.15 (1H, dd, *J* = 9.1, 9.2 Hz, 22 or 23-H), 5.03 (1H, dd, *J* = 8.6, 8.7 Hz, 22 or 23-H), 3.60 (1H, m, 3-H), 1.03 (3H, d, *J* = 6.5 Hz, 21-CH_3), 0.86 (3H, d, *J* = 6.0 Hz, 29-CH_3), 0.84 (6H, d, *J* = 6.6 Hz, 26-CH_3 and 27-CH_3), 0.80 (3H, s, 19-CH_3), 0.55 (3H, s, 18-CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 31.4 (t, C-1), 31.8 (t, C-2), 71.1 (d, C-3), 38.1 (t, C-4), 40.3 (d, C-5), 29.7 (t, C-6), 117.5 (d, C-7), 139.6 (s, C-8), 49.5 (d, C-9), 34.2 (s, C-10), 21.6 (t, C-11), 39.5 (t, C-12), 43.3 (s, C-13), 55.1 (d, C-14), 23.0 (t, C-15), 28.5 (t, C-16), 55.9 (d, C-17), 12.0 (q, C-18), 13.0 (q, C-19), 40.8 (d, C-20), 21.0 (q, C-21), 138.2 (d, C-22), 129.5 (d, C-23), 51.3 (d, C-24), 31.9 (d, C-25), 21.6 (q, C-26), 19.0 (q, C-27), 25.4 (t, C-28), 12.2 (q, C-29). The NMR data were identical to those of literature [5].

α-Spinasterol 3-O-β-D-glucopyranoside (2) Colorless needles (MeOH), $C_{35}H_{58}O_6$, mp. 275-277 °C, FAB-MS (negative) *m/z*: 573 $[M-H]^-$. ^{1}H NMR (MeOD, 500 MHz) δ: 5.30 (1H, dd, *J* = 8.7, 8.8 Hz, 22 or 23-H), 5.18 (1H, dd, *J* = 8.8, 8.7 Hz, 22 or 23-H), 4.88 (1H, d, *J* = 6.4 Hz, 1′-H), 4.57 (2H, d, *J* = 11.6 Hz, 6′-H), 4.39 (1H, t, *J* = 11.5 Hz, 4′-H), 4.01 (1H, m, 3-H), 1.00 (3H, d, *J* = 6.3 Hz, 21-CH_3), 0.92 (3H, d, *J* = 7.6 Hz, 26-CH_3), 0.89 (3H, t, *J* = 6.8 Hz, 29-CH_3), 0.88 (3H, d, *J* = 8.1 Hz, 27-CH_3), 0.73 (3H, s, 19-CH_3), 0.59 (3H, s, 18-CH_3); ^{13}C NMR (MeOD, 100 MHz) δ: 37.5 (t, C-1), 30.1 (t, C-2), 77.5 (d, C-3), 34.9 (t, C-4), 40.4

(d, C-5), 29.8 (t, C-6), 117.9 (d, C-7), 139.8 (s, C-8), 49.8 (d, C-9), 34.7 (s, C-10), 21.9 (t, C-11), 39.9 (t, C-12), 43.7 (s, C-13), 55.4 (d, C-14), 23.4 (t, C-15), 29.8 (t, C-16), 56.6 (d, C-17), 12.3 (q, C-18), 13.1 (q, C-19), 40.4 (d, C-20), 21.9 (q, C-21), 138.6 (d, C-22), 129.9 (d, C-23), 49.8 (d, C-24), 32.2 (d, C-25), 21.6 (q, C-26), 20.0 (q, C-27), 25.4 (t, C-28), 12.1 (q, C-29), 102.5 (d, C-1′), 75.4 (d, C-2′), 78.3 (d, C-3′), 72.0 (d, C-4′), 78.7 (d, C-5′), 63.2 (t, C-6′). The NMR data were consistent with those of reported [6].

β-Sitosterol (3) Colorless needles (EtOAc), $C_{29}H_{50}O$, mp. 143-144 ℃. It was confirmed by comparing it with the standard sample.

Stigmast-7-en-3-*O*-β-D-glucopyranoside (4) Colorless needles (MeOH), $C_{35}H_{60}O_6$, mp. 267-270 ℃, FAB-MS (negative) *m/z*: 575 ([M-H]$^-$, 100). ^{1}H NMR (pyridine-d_5, 500 MHz) δ: 5.03 (1H, d, *J* = 7.7 Hz, 1′-H), 4.59 (2H, d, *J* = 10.5 Hz, 6′-H), 4.38 (1H, t, *J* = 11.7 Hz, 4′-H), 4.01 (1H, m, 3-H), 0.99 (3H, d, *J* = 6.3 Hz, 21-CH_3), 0.90 (3H, d, *J* = 7.5 Hz, 26-CH_3), 0.88 (3H, t, *J* = 6.9 Hz, 29-CH_3), 0.86 (3H, d, *J* = 6.8 Hz, 27-CH_3), 0.72 (3H, s, 19-CH_3), 0.58 (3H, s, 18-CH_3); ^{13}C NMR (pyridine-d_5, 100 MHz) δ: 37.4 (t, C-1), 30.1 (t, C-2), 77.2 (d, C-3), 34.8 (t, C-4), 40.3 (d, C-5), 30.1 (t, C-6), 117.9 (d, C-7), 139.7 (s, C-8), 49.7 (d, C-9), 34.6 (s, C-10), 21.8 (t, C-11), 39.9 (t, C-12), 43.7 (s, C-13), 55.3 (d, C-14), 23.5 (t, C-15), 28.3 (t, C-16), 56.4 (d, C-17), 12.1 (q, C-18), 13.1 (q, C-19), 36.9 (d, C-20), 19.2 (q, C-21), 34.5 (t, C-22), 28.3 (t, C-23), 46.2 (d, C-24), 29.6 (d, C-25), 19.3 (q, C-26), 20.0 (q, C-27), 23.4 (t, C-28), 12.2 (q, C-29), 102.4 (d, C-1′), 75.4 (d, C-2′), 78.7 (d, C-3′), 71.9 (d, C-4′), 78.5 (d, C-5′), 63.0 (t, C-6′). The NMR data were equal to those of literature [6].

22-Methylene-9, 19-cyclolanostan-3β-ol (5) White plates (MeOH), $C_{31}H_{52}O$, mp. 124-125 ℃, EI-MS *m/z*: 440 (25), 315 (15), 300 (50), 175 (90). ^{1}H NMR ($CDCl_3$, 500 MHz) δ: 4.66 (2H, d, *J* = 5.0 Hz, 31-CH_2), 3.26 (1H, m, 3-H), 1.25 (3H, s, 26-H), 1.25 (3H, s, 27-H), 1.03 (3H, d, *J* = 6.8 Hz, 21-CH_3), 0.96 (3H, s, 29-CH_3), 0.89 (3H, s, 18-CH_3), 0.88 (3H, s, 28-CH_3), 0.80 (3H, s, 30-CH_3), 0.54, 0.32 (each 1H, d, *J* = 5.0 Hz, 19-CH_2); ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 31.7 (t, C-1), 29.6 (t, C-2), 78.5 (d, C-3), 40.8 (s, C-4), 47.0 (d, C-5), 21.1 (t, C-6), 27.9 (t, C-7), 47.8 (d, C-8), 19.7 (s, C-9), 26.0 (s, C-10), 26.0 (t, C-11), 35.5 (t, C-12), 45.1 (s, C-13), 48.5 (s, C-14), 32.7 (t, C-15), 26.8 (t, C-16), 52.1 (d, C-17), 17.9 (q, C-18), 29.6 (t, C-19), 36.9 (d, C-20), 18.3 (q, C-21), 158.3 (s, C-22), 33.4 (t, C-23), 35.5 (t, C-24), 31.1 (d, C-25), 21.0 (q, C-26), 22.5 (q, C-27), 19.5 (q, C-28), 25.4 (q, C-29), 15.4 (q, C-30), 107.3 (t, C-31). The NMR data were identical to those of literature [7].

(2S, 3S, 4R, 10E)-2-(2′, 3′-Dihydroxytetracosanoylamino)-10-octadecene-1, 3, 4-triol (6) White powder (MeOH), $C_{42}H_{83}O_6N$, FAB-MS (negative) *m/z*: 696 [M-H]$^-$. ^{1}H NMR (pyridine-d_5, 500 MHz) δ: 8.68 (1H, d, *J* = 9.3 Hz, NH), 5.53 (1H, m, H-10), 5.50 (1H, m, H-11), 5.12 (1H, m, H-2), 4.76 (1H, m, H-2′), 4.55 (1H, m, H-3′), 4.54 (1H, dd, *J* = 8.5, 4.2 Hz, H-1), 4.43 (1H, dd, *J* = 8.5, 4.2 Hz, H-1), 4.33 (1H, m, H-3), 4.28 (1H, m, H-4), 1.98-2.06 (m), 1.25-1.31 (54H, m, 27 × CH_2), 0.86 (6H, t-like, *J* = 6.6 Hz, Me-18 and Me-24′); ^{13}C NMR (pyridine-d_5, 100 MHz) δ: 62.0 (t, C-1), 53.1 (d, C-2), 76.3 (d, C-3), 72.9 (d, C-4), 33.9 (t, C-5), 26.6 (t, C-6), 33.0 (t, C-7), 33.4 (t, C-8), 32.2 (t, C-9), 130.8 (d, C-10), 130.7 (d, C-11), 32.6 (t, C-12), 174.0 (s, C-1′), 76.8 (d, C-2′), 73.7 (d, C-3′), 26.6 (t, C-4′), 29.6-30.3 (t, C-13-16 and C-5′-22′), 23.0 (t, C-17 and C-23′), 14.3 (q, C-18, 24′). The NMR data resembled those of literature [8].

(2S, 3S, 4R, 10E)-2-[(2′R)-2-Hydroxytetracosanoylamino]-10-octadecene-1, 3, 4-triol (7) White powder (MeOH); mp. 138-139 ℃; $C_{42}H_{83}O_5N$; FAB-MS (negative) *m/z*: 680 ([M-H]$^-$); ^{1}H NMR (pyridine-d_5, 500 MHz) δ: 8.57 (1H, d, *J* = 8.8 Hz, NH), 5.52 (1H, m, H-10), 5.50 (1H, m, H-11), 5.10 (1H, d, *J* = 4.3 Hz, H-2), 4.61 (1H, m, H-2′), 4.51 (1H, *br*s, H-1), 4.42 (1H, m, H-1), 4.33 (1H, m, H-3), 4.27 (1H, m, H-4), 1.25-1.30 (54H, m, 27

×CH_2), 0.85 (6H, t-like, J = 6.7 Hz, Me-18 and Me-24′); ^{13}C NMR (pyridine-d_5, 100 MHz) δ: 62.0 (t, C-1), 52.9 (d, C-2), 76.9 (d, C-3), 73.0 (d, C-4), 33.3 (t, C-5), 26.8 (t, C-6), 32.2 (t, C-9), 130.8 (d, C-10), 130.7 (d, C-11), 33.0 (t, C-12), 175.3 (s, C-1′), 72.5 (d, C-2′), 35.7 (t, C-3′), 25.8 (t, C-4′), 29.6-30.3 (t, C-13-16 and C-5′-22′), 22.9 (t, C-17 and C-23′), 14.3 (q, C-18, 24′). The NMR data were identical to those of literature [9].

(2S, 3S, 4R, 10E)-1-(β-D-Glucopyranosyl)-2-[(2′R)-2′-hydroxytetracosanoylamino]-10-octadecene-1, 3, 4-triol (8) White powder (MeOH); $C_{48}H_{93}O_{10}N$; FAB-MS (negative) m/z, 842 ([M-H]$^-$, 100), 680 (15). ^{1}H NMR (pyridine-d_5, 500 MHz) δ: 8.54 (1H, d, J = 8.8 Hz, NH), 5.50 (1H, m, H-10), 5.49 (1H, m, H-11), 5.27 (1H, d, J = 4.3 Hz, H-2), 4.93 (1H, d, J = 6.9 Hz, H-1″), 1.24-1.31 (54H, m, 27 × CH_2), 0.85 (6H, t-like, J = 6.7 Hz, Me-18 and Me-24′); ^{13}C NMR (pyridine-d_5, 100 MHz) δ: 70.4 (t, C-1), 51.9 (d, C-2), 75.9 (d, C-3), 72.5 (d, C-4), 33.9 (t, C-5), 33.0 (t, C-6), 32.9 (t, C-7), 32.8 (t, C-8), 32.2 (t, C-9), 130.9 (d, C-10), 130.7 (d, C-11), 33.3 (t, C-12), 175.7 (s, C-1′), 72.5 (d, C-2′), 35.6 (t, C-3′), 25.9 (t, C-4′), 29.6-30.0 (t, C-13-16 and C-5′-22′), 23.0 (t, C-17 and C-23′), 14.3 (q, C-18, 24′); Glc: 105.5 (d, C-1″), 75.2 (d, C-2″), 78.5 (d, C-3″), 71.6 (d, C-4″), 78.6 (d, C-5″), 62.7 (t, C-6″). The NMR data were equal to those of literature [10].

Soya-cerbroside I (9) White powder (MeOH); $C_{40}H_{75}O_9N$; FAB-MS (negative) m/z, 712 ([M-H]$^-$, 100), 550 (15). ^{1}H NMR (pyridine-d_5, 400 MHz) δ: 8.38 (1H, d, J = 8.8 Hz, NH), 5.77 (1H, m, H-5), 5.49 (3H, m, H-4, H-8 and H-9), 4.92 (1H, d, J = 7.7 Hz, H-1″), 4.51 (1H, m, H-6″b), 4.38 (1H, m, H-6″a), 4.25 (2H, m, H-1a and H-3″), 4.21 (1H, m, H-4″), 4.05 (4H, m, H-1b, H-2, H-3 and H-2″), 3.92 (2H, m, H-2′ and H-5″), 2.14 (4H, *brs*, H-6, H-7), 1.99 (2H, m, H-10), 1.71 (1H, m, H-3′), 1.37 (1H, m, H-4′), 1.25-1.35 (38H, m, 19 ×CH_2), 0.86 (6H, t-like, J = 6.9 Hz, H-18 and H-16′); ^{13}C NMR (pyridine-d_5, 100 MHz) δ: 70.2 (t, C-1), 54.6 (d, C-2), 71.5 (d, C-3), 131.1 (d, C-4), 132.1 (d, C-5), 32.2 (t, C-6), 32.1 (t, C-7), 130.0 (d, C-8), 132.1 (d, C-9), 32.9 (d, C-10), 175.7 (s, C-1′), 72.3 (d, C-2′), 35.7 (t, C-3′), 25.9 (t, C-4′), 29.6-30.0 (t, C-11-16 and C-5′-14′), 23.0 (t, C-17 and C-15′), 14.3 (q, C-18, 16′); Glc: 105.7 (d, C-1″), 75.2 (d, C-2″), 78.5 (d, C-3″), 71.5 (d, C-4″), 78.6 (d, C-5″), 62.7 (t, C-6″). The NMR data resembled those of literature [9]. In summary, nine compounds were isolated from the chloroform fraction of the methanol extract of *C. sativus* L. stems. They were elucidated by physicochemical properties and spectroscopic analysis except compound 3 which was determined to be β-sitosterol by comparing with its TLC behavior with standard sample. By literature study, the compounds except β-sitosterol were isolated from this plant for the first time.

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