云南黄连中非生物碱类化学成分的研究

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摘 要 对黄连属植物云南黄连 *Coptis teeta* wall. 根茎的乙醇提取物进行反复硅胶、凝胶、反相柱色谱和制备型高效液相色谱分离纯化,运用波谱学方法鉴定了 12 个化合物,分别为 3,5,7-三羟基-6,8-二甲基黄酮(3,5,7-trihydroxy-6,8-dimethylflavone,**1**),阿魏酸(ferulic acid,**2**),*Z*-咖啡酸硬脂醇酯(*Z*-octadecyl caffeate,**3**),原儿茶酸(protocatechuic acid,**4**),丹参素甲酯(methyl-3,4-dihydroxyphenyl lactate,**5**),3,4-二羟基苯乙醇(3,4-dihydroxy phenethyl alcohol,**6**),3,5-dihydroxyphenethyl alcohol-3-*O*-*β*-D-glucopyranoside(**7**),(+)-落叶松树脂醇[(+)-lariciresinol,**8**],woorenoside Ⅰ(**9**),woorenoside Ⅱ(**10**),longifolroside A(**11**),(+)-syringaresinol-4-*O*-*β*-D-glucopyranoside(**12**)。所有化合物均为首次从云南黄连中得到,其中化合物 **1**,**4**,**7**,**11** 首次从该属植物中得到。

关键词 云南黄连;非生物碱类;化学成分;结构鉴定

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Non-alkaloid chemical constituents from the rhizome of *Coptis teeta*

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Abstract Twelve compounds were isolated and purified from the ethanol extract of *Coptis teeta* wall by various chromatographic methods, and their structures were elucidated by spectral techniques and physicochemical properties as 3, 5, 7-trihydroxy- 6, 8-dimethylflavone(**1**), ferulic acid(**2**), *Z*-octadecyl caffeate(**3**), protocatechuic acid (**4**), methyl-3, 4-dihydroxyphenyl lactate(**5**), 3, 4-dihydroxyphenethyl alcohol(**6**), 3, 5-dihydroxyphenethyl alcohol-3-*O*-*β*-D-glucopyranoside(**7**), (+)-lariciresinol(**8**), woorenoside I (**9**), woorenoside II (**10**), longifolroside A (**11**) and (+)-syringaresinol-4-*O*-*β*-D-glucopyranoside(**12**). Compounds **1**, **4**, **7**, **11** were isolated from this genus for the first time. All the compounds were isolated from this plant for the first time.

Key words *Coptis teeta*; non-alkaloid; chemical constituents; structural identification

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黄连是我国常用著名中药,味苦、性寒,具有清热燥湿、泻火解毒等功效[1]。《中华人民共和国药典》(2010 版)中规定其植物来源为黄连(*Coptis chinensis* Franch)、云南黄连(*C. teeta* Wall)和三角叶黄连(*C. deltoidea* C. Y. Cheng et Hsiao)的干燥根茎。现代药理研究表明,黄连中化学成分具有抗肿瘤、降血糖、降血脂、抗菌、心脑血管保护等多种作用[2]。目前已有的研究报道集中在黄连的生物碱类化学成分,而云南黄连化学成分的研究未见报道。前期预实验发现,云南黄连中的生物碱类成分与其他两种植物中的类似,但非生物碱类成分存在一定差异。为阐明云南黄连中非生物碱类成分,本研究采用各种色谱方法和光谱技术,从云南黄连干燥根茎的乙醇提取物中分离并鉴定 12 个化合物,包括 1 个黄酮:3,5,7-三羟基-6,8-二甲基黄酮(3,5,7-trihydroxy-6,8-dimethylflavone,**1**);6 个苯环类化合物:阿魏酸(ferulic acid,**2**),*Z*-咖啡酸硬脂醇酯(*Z*-octadecyl caffeate,**3**),原儿茶酸(protocatechuic acid,**4**),丹参素甲酯(methyl-3,4-dihydroxyphenyl lactate,**5**),3,4-二羟基苯乙醇(3,4-dihydroxyphenethyl alcohol,**6**),3,5-dihydroxyphenethyl alcohol-3-*O*-*β*-D- glucopyranoside(**7**);5 个木脂素:(+)-落叶松树脂醇[(+)-lariciresinol,**8**],woorenoside Ⅰ(**9**),woorenoside Ⅱ(**10**),longifolroside A(**11**),(+)-syringaresinol-4-*O*-*β*-D-glucopyranoside(**12**)。以上化合物均首次从云南黄连中分离得到,其中化合物 **1**,**4**,**7**,**11** 首次从该属植物中得到。

1 材 料

X-4 型数字显示双目显微熔点测定仪(温度未校正);核磁共振波谱用德国 Bruker 公司 Avance-300(^{1}H NMR 300 MHz,^{13}C NMR 75 MHz)和 Bruker Avance-500(^{1}H NMR 500 MHz,^{13}C NMR 125 MHz)测定;安捷伦公司 HP-1100 LC/EST 型液质联用仪。Pre-HPLC(美国 AllTech 公司);薄层色谱硅胶 GF_{254};柱色谱硅胶(青岛海洋化工厂);Sephadex LH-20(美国 Pharmacia 公司);ODS C_{18}柱(美国 Merck 公司);其余试剂均为市售分析纯。

云南黄连药材采自云南大理,经中国药科大学生药学教研室张勉教授鉴定为云南黄连 *Coptis teeta* wall 根茎,标本保存于中国药科大学中药学院天然药物化学教研室。

2 提取和分离

云南黄连干燥根茎 19 kg,用 80% 乙醇热回流提取两次,70% 乙醇热回流提取 1 次,合并提取液,回收溶剂得浸膏,浸膏用水混悬,依次用石油醚、乙酸乙酯萃取。乙酸乙酯部位 115 g,经硅胶、凝胶、ODS、制备型液相色谱得化合物 **1**(20 mg)、**2**(10 mg)、**3**(82 mg)、**4**(35 mg)、**5**(3 mg)、**6**(3 mg)、**8**(100 mg)、**9**(15 mg)、**10**(10 mg)、**11**(3 mg)。取水部位 900 g,经硅胶、凝胶、ODS、制备型液相色谱得化合物 **7**(5 mg)和 **12**(6 mg)。

3 结构鉴定

化合物 **1** 黄色针状晶体(氯仿),mp:170 ~ 172 ℃,溶于氯仿。^{1}H NMR(DMSO-d_6,300 MHz) δ:12.51(1H,s,5-OH),9.72(1H,s,7-OH),9.59(1H,s,3-OH),8.19(2H,d,J = 8.0 Hz,H-2′,6′),7.48 ~ 7.60(3H,m,H-3′,4′,5′),2.29(3H,s,8-CH_3),2.08(3H,s,6-CH_3)。^{13}C NMR(75 MHz) δ:176.4(C-4),159.8(C-7),155.1(C-5),151.7(C-9),145.4(C-2),136.9(C-3),131.3(C-1′),129.8(C-4′),128.6(C-2′,6′),127.4(C-3′,5′),106.5(C-6),103.1(C-8),101.5(C-10),8.1(8-CH_3),8.0(6-CH_3)。以上波谱数据与文献[3]对照一致,确定化合物为 3,5,7-三羟基-6,8-二甲基黄酮(3,5,7-trihydroxy-6,8-dimethylflavone)。

化合物 **2** 黄色针状晶体(氯仿),mp:170 ~ 172 ℃,溶于氯仿。^{1}H NMR(DMSO-d_6,300 MHz) δ:12.10(1H,s,-COOH),9.53(1H,s,-OH),7.49(1H,d,J = 15.9 Hz,H-7),7.28(1H,d,J = 1.8 Hz,H-2),7.08(1H,dd,J = 8.2,1.8 Hz,H-6),6.79(1H,d,J = 8.2 Hz,H-5),6.36(1H,d,J = 15.9 Hz,H-8),3.82(3H,s,-OCH_3)。^{13}C NMR(75 MHz) δ:167.9(C-9),149.0(C-3),147.9(C-4),144.4(C-7),125.7(C-1),122.8(C-8),115.6(C-6),115.5(C-5),111.1(C-2),55.7(-OCH_3)。以上波谱数据与文献[4]对照一致,确定化合物为阿魏酸(ferulic acid)。

化合物 **3** 白色固体,mp:92 ~ 93 ℃,溶于氯仿、甲醇。ESI-MS m/z 431.3[M − H]$^-$,178.8[M − $C_{18}H_{37}$]$^-$,433.2[M + H]$^+$。^{1}H NMR(CD_3OD,300 MHz) δ:7.53(1H,d,J = 15.9 Hz,H-8),7.04(1H,d,J = 2.0 Hz,H-2),6.94(1H,dd,J = 8.3,2.0 Hz,H-6),6.78(1H,d,J = 8.3 Hz,H-5),6.27(1H,d,J = 15.9 Hz,H-7),4.17(2H,t,J = 6.6 Hz,H-1′),1.69(2H,m,J = 6.8 Hz,H-2′),1.28(30H,br. s,CH_2 × 15),0.90(3H,t,J = 6.9 Hz,H-18′)。以上波谱数据与文献[5]对照一致,确定化合物为 *Z*-咖啡酸硬脂醇酯(*Z*-octadecyl caffeate)。

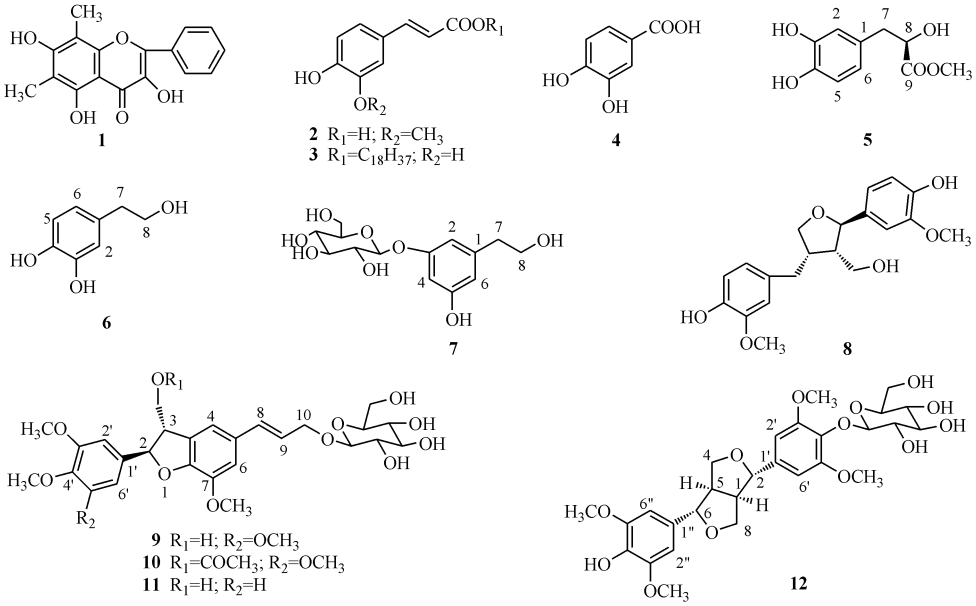

Figure 1　Chemical structures of compounds **1-12**

化合物 **4**　淡黄色针晶(甲醇),mp:197 ~ 198 ℃,溶于甲醇。紫外灯 254 nm 下观察有暗斑,香草醛-浓硫酸不显色。与原儿茶酸对照品在 HPTLC 中的 R_f 及显色行为一致,且两者混合熔点不下降,确定化合物为原儿茶酸(protocatechuic acid)。

化合物 **5**　淡黄色固体,mp:146 ~ 148 ℃溶于二氯甲烷、氯仿。^{1}H NMR(DMSO-d_6,300 MHz) δ:8.70(1H,s,4-OH),8.61(1H,s,3-OH),6.60(1H,d,J = 7.9 Hz,H-5),6.58(1H,d,J = 1.9 Hz,H-2),6.42(1H,dd,J = 8.0,2.0 Hz,H-6),5.43(1H,br. s,8-OH),4.11(1H,t,J = 6.2 Hz,H-8),3.59(3H,s,-OCH_3),2.75(1H,dd,J = 5.4,13.6 Hz,H-7a),2.63(1H,dd,J = 7.9,13.7 Hz,H-7b)。^{13}C NMR(75 MHz) δ:174.0(C-9),144.7(C-3),143.7(C-4),128.2(C-1),119.9(C-6),116.7(C-2),115.2(C-5),71.6(C-8),51.2(C-OCH_3),39.6(C-7)。以上波谱数据与文献[6]对照一致,确定化合物为丹参素甲酯(methyl-3,4-dihydroxyphenyl lactate)。

化合物 **6**　黄色油状物,溶于氯仿、甲醇。^{1}H NMR(C_5D_5N,300 MHz) δ:7.32(1H,d,J = 2.0 Hz,H-2),7.49(1H,d,J = 7.8 Hz,H-5),6.88(1H,dd,J = 8.0,2.1 Hz,H-6),4.11(2H,t,J = 7.1 Hz,H-8),3.04(2H,t,J = 7.1 Hz,H-7)。^{13}C NMR(75 MHz) δ:147.9(C-3),146.3(C-4),132.4(C-1),121.4(C-6),118.4(C-2),117.3(C-5),64.8(C-8),40.8(C-7)。以上波谱数据与文献[7]对照一致,确定化合物为 3,4-二羟基苯乙醇(3,4-dihydroxyphenethyl alcohol)。

化合物 **7**　白色固体,mp:104 ~ 106 ℃,溶于甲醇。^{1}H NMR(DMSO-d_6,300 MHz) δ:6.95(1H,br. s,H-4),6.70(1H,br. s,H-2),6.69(1H,br. s,H-6),4.63(1H,d,J = 7.4 Hz,H-glc-1),3.53(2H,t,J = 7.3 Hz,H-8),2.59(2H,t,J = 7.3 Hz,H-7)。^{13}C NMR(75 MHz) δ:145.0(C-3),145.0(C-5),130.3(C-1),123.2(C-2),117.6(C-6),115.5(C-4),102.5(C-glc-1),77.2(C-glc-3),75.9(C-glc-5),73.4(C-glc-2),69.8(C-glc-4),60.8(C-glc-6),62.3(C-8),38.5(C-7)。以上波谱数据与文献[8]对照一致,确定化合物为 3,5-dihydroxyphenethyl alcohol-3-*O*-β-D-glucopyranoside。

化合物 **8**　淡黄色固体,mp:170 ~ 172 ℃,溶于氯仿、甲醇。^{1}H NMR(DMSO-d_6,300 MHz) δ:8.81(1H,s,4′-OH),8.68(1H,s,4-OH),6.82(1H,d,J = 0.9 Hz,H-2′),6.74(1H,d,J = 1.7 Hz,H-2),6.69(1H,br. d,J = 8.0 Hz,H-6′),6.67(2H,d,J = 8.0 Hz,H-5,5′),6.57(1H,dd,J = 8.0,1.8 Hz,H-6),4.67(1H,t,J = 4.6 Hz,9′-OH),4.65(1H,d,J = 6.2 Hz,H-7′),3.87(1H,dd,J = 8.0,6.6 Hz,H-9b),3.74(3H,s,3-OCH_3)、3.74(3H,s,3′-OCH_3),3.66(1H,m,H-9′a),3.55(1H,dd,J = 7.8,6.6 Hz,H-9a),3.46(1H,m,H-9′b),2.82(1H,dd,J = 13.2,4.6 Hz,H-7b),2.59(1H,m,H-8),2.41(1H,dd,J = 13.0,11.1 Hz,H-7a),2.18(1H,ddt,J = 6.8,6.8,6.8 Hz,H-8′)。^{13}C NMR(75 MHz) δ:147.5(C-3′),147.4(C-3),145.5(C-4′),144.6(C-4),134.7(C-1′),131.8(C-1),120.6(C-6),118.2(C-6′),115.4(C-5′),115.1(C-5),112.8(C-2),110.0(C-2′),81.8(C-7′),71.8(C-9),58.6(C-9′),55.6(3-OCH_3),55.6(3′-OCH_3),52.4(C-8′),42.0(C-8),32.2(C-7)。以上波谱数据与文献[5]对照一致,确定化合物为(+)-落叶松树脂醇[(+)-lariciresinol]。

化合物 **9**　白色固体(甲醇),mp:172 ~ 173 ℃,溶于甲醇。^{1}H NMR(DMSO-d_6,500 MHz) δ:6.97(2H,s,H-4,6),6.67(2H,s,H-2′,6′),6.58(1H,d,J = 15.9 Hz,H-8),6.21(1H,dt,J = 16.0,6.0 Hz,H-9),5.51(1H,d,J = 6.8 Hz,H-2),4.48(1H,t,J = 6.0 Hz,3-CH_2OH),4.40(1H,m,H-

10a),4.21(1H,d,J = 5.8 Hz,H-glc-1),4.19(1H,m,H-10b),3.82(3H,s,OCH_3),3.75(6H,s,OCH_3),3.65(3H,s,OCH_3)。^{13}C NMR(125 MHz) δ:152.9(C-3′),152.9(C-5′),147.3(C-7a),143.7(C-7),137.1(C-1′),137.0(C-4′),131.7(C-8),130.3(C-5),129.4(C-3a),123.7(C-9),115.2(C-4),110.6(C-6),103.2(C-2′),103.2(C-6′),102.0(C-glc-1),87.1(C-2),76.9(C-glc-3),76.8(C-glc-5),73.5(C-glc-2),70.1(C-glc-4),68.7(C-10),62.8(3-CH_2OH),61.1(C-glc-6),60.0,55.9,55.9,55.8(7,3′,4′,5′-OCH_3),53.1(C-3)。以上波谱数据与文献[9]对照一致,确定化合物为 woorenoside Ⅰ。

化合物 **10** 白色固体(甲醇),mp:141 ~ 142 ℃,溶于甲醇。ESI-MS m/z 624[M + NH_4]$^+$,641[M + Cl]$^-$,分子式为 $C_{30}H_{38}O_{13}$。^{1}H NMR(DMSO-d_6,300 MHz) δ:7.02(1H,s,H-4),6.99(1H,s,H-6),6.70(2H,s,H-2′,5′),6.60(1H,d,J = 15.9 Hz,H-8),6.25(1H,dt,J = 15.8,5.6 Hz,H-9),5.49(1H,d,J = 7.6 Hz,H-2),4.22(1H,d,J = 7.7 Hz,H-glc-1),3.83(3H,s),3.76(6H,s),3.66(3H,s),(7,3′,4′,5′-OCH_3),2.00(3H,s,-$COCH_3$)。^{13}C NMR(75 MHz) δ:170.3(-CO-),153.0(C-3′),153.0(C-5′),147.3(C-7a),143.9(C-7),137.4(C-4′),136.1(C-1′),131.6(C-8),130.7(C-5),128.0(C-3a),124.0(C-9),115.0(C-4),110.9(C-6),103.5(C-2′),103.5(C-6′),102.0(C-glc-1),87.5(C-2),76.9(C-glc-3),76.8(C-glc-5),73.5(C-glc-2),70.1(C-glc-4),68.7(C-10),64.7(C-CH_2OH),61.1(C-glc-6),60.0,55.9,55.9,55.8(7,3′,4′,5′-OCH_3),49.4(C-3),20.6(-CH_3)。以上波谱数据与文献[9]对照一致,确定化合物为 woorenoside Ⅱ。

化合物 **11** 白色固体(甲醇),mp:215 ~ 216 ℃,溶于甲醇。^{1}H NMR(DMSO-d_6,500 MHz) δ:6.97(1H,br. s,H-4),6.96(1H,br. s,H-2′),6.95(1H,br. s,H-6),6.93(1H,d,J = 7.0 Hz,H-5′),6.88(1H,dd,J = 8.3,1.7 Hz,H-6′),6.57(1H,d,J = 15.9 Hz,H-8),6.21(1H,dt,J = 15.9,6.0 Hz,H-9),5.51(1H,d,J = 6.7 Hz,H-2),4.40(1H,m,H-10a),4.21(1H,d,J = 5.8 Hz,H-glc-1),4.18(1H,m,H-10b),3.81,3.74,3.73(each 3H,s,OCH_3)。^{13}C NMR(125 MHz) δ:148.8(C-4′),148.6(C-3′),147.4(C-7a),143.7(C-7),133.9(C-1′),131.8(C-8),130.2(C-5),129.4(C-3a),123.6(C-9),118.1(C-6′),115.2(C-4),111.8(C-5′),110.5(C-6),109.8(C-2′),102.0(C-glc-1),87.0(C-2),76.9(C-glc-3),76.8(C-glc-5),73.5(C-glc-2),70.1(C-glc-4),68.7(C-10),62.9(3-CH_2OH),61.1(C-glc-6),55.7,55.6,55.5(7,3′,4′-OCH_3),53.0(C-3)。以上波谱数据与文献[10]对照一致,确定化合物为 longifloroside A。

化合物 **12** 白色固体,mp:187 ~ 188 ℃,溶于甲醇。^{1}H NMR(DMSO-d_6,300 MHz) δ:8.25(1H,s,4″-OH),6.66(2H,s,H-2″,6″),6.60(2H,s,H-2′,6′),4.88(1H,d,J = 7.3 Hz,H-glc-1),4.67(1H,d,J = 4.2 Hz,H-2),4.64(1H,d,J = 4.2 Hz,H-6),4.28(1H,t,J = 5.0 Hz,H-4a),4.18(1H,t,J = 6.9 Hz,H-8a),3.80(1H,br. d,J = 3.8 Hz,H-4b),3.77(1H,br. d,J = 6.1 Hz,H-8b)、3.17-3.20(2H,m,H-1,5),3.76(6H,s,2 × OCH_3),3.75(6H,s,2 × OCH_3)。^{13}C NMR(75 MHz) δ:152.6(C-3″,5″),147.9(C-3′,5′),137.2(C-4′),134.8(C-4″),133.7(C-1″),131.3(C-1′),104.2(C-2″),104.2(C-6″),103.7(C-2′),103.7(C-6′),102.6(C-glc-1),85.3(C-2),85.0(C-6),77.2(C-glc-5),76.5(C-glc-3),74.1(C-glc-2),71.2(C-4),71.1(C-8),69.9(C-glc-4),60.9(C-glc-6),56.4(3″,5″-OCH_3),56.0(3′,5′-OCH_3),53.6(C-5),53.6(C-1)。以上波谱数据与文献[11]对照一致,确定化合物为(+)-syringaresinol-4-O-β-D-glucopyranoside。

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