乌拉尔甘草皂苷类成分研究

陶伟伟[1, 2, 3]，段金廒[1, 2, 3*]，杨念云[2, 3]，李建萍[2, 3]，唐于平[2, 3]，严　辉[2, 3]

1. 中国药科大学中药学院，江苏 南京　210009
2. 南京中医药大学 江苏省中药资源产业化过程协同创新中心，江苏 南京　210023
3. 南京中医药大学 江苏省方剂高技术研究重点实验室，江苏 南京　210023

摘　要：目的　研究乌拉尔甘草 *Glycyrrhiza uralensis* 根及根茎的化学成分。**方法**　应用溶剂法和色谱法进行分离纯化，利用波谱技术鉴定化合物结构；并测试化合物的细胞毒活性。**结果**　从乌拉尔甘草 50%乙醇提取物中分离得到 14 个皂苷类化合物，分别鉴定为 uralsaponin C（**1**）、uralsaponin D（**2**）、licorice-saponin A3（**3**）、uralsaponin F（**4**）、22β-acetoxyl-glycyrrizin（**5**）、24-hydroxyl-licorice-saponin E2（**6**）、licorice-saponin E2（**7**）、licorice-saponin G2（**8**）、22β-acetoxyl-glyrrhaldehyde（**9**）、3β-*O*-[β-*D*-glucuronopyranosyl-(1→2)-β-*D*-glucuronopyranosyl]-glycyrretol（**10**）、araboglycyrrhizin（**11**）、licorice-saponin J2（**12**）、甘草酸（**13**）、单葡糖醛酸基甘草次酸（**14**）。化合物 **1**～**14** 对 3 种人源肿瘤细胞 MGC-803、SW620、SMMC-7721 的半数抑制率（IC_{50}）均大于 100 μmol/L，化合物 **2**、**6**～**8**、**13** 的水解后苷元对 3 种人源肿瘤细胞的抑制率为 18.3～41.6 μmol/L。**结论**　化合物 **14** 是一个新的天然产物，化合物 **11** 为首次从该植物中分离得到；化合物 **1**～**14** 对 3 种人源肿瘤细胞 MGC-803、SW620、SMMC-7721 均无显著的细胞毒活性，化合物 **2**、**6**～**8**、**13** 水解后苷元细胞毒活性增强。

关键词：乌拉尔甘草；皂苷；甘草酸；单葡糖醛酸基甘草次酸；细胞毒活性

中图分类号：R284.1　　**文献标志码：**A　　**文章编号：**0253 - 2670(2013)12 - 1552 - 06

DOI: 10.7501/j.issn.0253-2670.2013.12.006

Chemical constituents of triterpenoid saponins from *Glycyrrhiza uralensis*

TAO Wei-wei[1, 2, 3], DUAN Jin-ao[1, 2, 3], YANG Nian-yun[2, 3], LI Jian-ping[2, 3], TANG Yu-ping[2, 3], YAN Hui[2, 3]

1. School of Traditional Chinese Medicine, China Pharmaceutical University, Nanjing 210009, China
2. Collaborative Innovation Center of Chinese Medicinal Resources Industrialization of Jiangsu Province, Nanjing University of Chinese Medicine, Nanjing 210023, China
3. Jiangsu Key Laboratory for High Technology Research of TCM Formulae, Nanjing University of Chinese Medicine, Nanjing 210023, China

Abstract: Objective　To study the chemical constituents from the roots and rhizomes of *Glycyrrhiza uralensis*. **Methods**　The compounds were separated and purified by solvent and chromatographic methods. Their structures were identified by spectroscopic techniques. **Results**　Fourteen triterpenoid saponins isolated from 50% ethanol extract of the roots and rhizomes of *G. uralensis* were identified as uralsaponin C (**1**), uralsaponin D (**2**), licorice-saponin A3 (**3**), uralsaponin F (**4**), 22β-acetoxyl-glycyrrizin (**5**), 24-hydroxyl-licorice-saponin E2 (**6**), licorice-saponin E2 (**7**), licorice-saponin G2 (**8**), 22β-acetoxyl-glyrrhaldehyde (**9**), 3β-*O*-[β-*D*-glucuronopyranosyl-(1→2)-β-*D*-glucuronopyranosyl]-glycyrretol (**10**), araboglycyrrhizin (**11**), licorice-saponin J2 (**12**), glycyrrhizin (**13**), and glycyrrhetic acid monoglucuronide (**14**). Compounds **1**—**14** showed the cytotoxic activity against the human cancer cell lines MGC-803, SW620, and SMMC-7721 with IC_{50} > 100 μmol/L. The aglycones of compounds **2**, **6**—**8**, and **13** displayed the inhibition on the growth of cancer cells with IC_{50} at 18.3—41.6 μmol/L. **Conclusion**　Compound **14** is a new natural product, and compound **11** is isolated from the plant for the first time; Compounds **1**—**14** show no cytotoxic activity against the human cancer cell lines MGC-803, SW620, and SMMC-7721, and the aglycones of compounds **2**, **6**—**8**, and **13** could significantly increase the cytotoxic activity after hydrolysis.

Key words: *Glycyrrhiza uralensis* Fisch.; saponins; glycyrrhizin; glycyrrhetic acid monoglucuronide; cytotoxic activity

甘草为豆科植物乌拉尔甘草 *Glycyrrhiza uralensis* Fisch.、胀果甘草 *G. inflata* Bat.、光果甘草 *G. glabra* L. 的干燥根及根茎。其味甘，性平，归心、肺、脾、胃经。具有补脾益气、清热解毒、祛痰止

收稿日期：2012-12-21

基金项目：国家重点基础研究发展计划（“973”计划）（2011CB505300，2011CB505303）；江苏高校优势学科建设工程资助项目（ysxk-2010）

作者简介：陶伟伟（1984—），男，博士研究生，研究方向为方剂配伍及物质基础研究。E-mail: tw845@163.com

***通信作者**　段金廒，教授，博士生导师。Tel: (025)85811116　E-mail: dja@njutcm.edu.cn

咳、缓急止痛、调和诸药的功效。药理研究表明，甘草具有抗氧化、抗病毒、抗抑郁、抗炎、抗肾炎以及保肝活性[1-2]。甘草中三萜皂苷是其主要的活性物质基础，主要由 1 分子的五环三萜苷元与 2 分子的葡萄糖醛酸构成。相关研究表明，这些主要的皂苷类成分具有保肝、抗癌、抗炎、抗病毒以及神经保护活性等作用[3-4]。本实验对甘草中皂苷类成分进行了系统分离并得到 14 个化合物，分别鉴定为 uralsaponin C（**1**）、uralsaponin D（**2**）、licorice-saponin A3（**3**）、uralsaponin F（**4**）、22β-acetoxyl-glycyrrizin（**5**）、24-hydroxyl-licorice-saponin E2（**6**）、licorice-saponin E2（**7**）、licorice-saponin G2（**8**）、22β-acetoxyl-glyrrhaldehyde（**9**）、3β-*O*-[β-*D*-glucuronopyranosyl-(1→2)-β-*D*-glucuronopyranosyl]-glycyrretol（**10**）、araboglycyrrhizin（**11**）、licorice-saponin J2（**12**）、甘草酸（glycyrrhizin，**13**）、单葡糖醛酸基甘草次酸（glycyrrhetic acid monoglucuronide，**14**）。化合物 **14** 是一个新的天然产物，化合物 **11** 为首次从该植物中分离得到；化合物 **1**～**14** 对 3 种人源肿瘤细胞 MGC-803、SW620、SMMC-7721 均无显著的细胞毒活性，化合物 **2**、**6**～**8**、**13** 水解后苷元细胞毒活性增强。

1 仪器与材料

Bruker Avance AV 500/300 型核磁共振仪（德国 Bruker 公司）；Waters Synapt^{TM} Q—TOF 型质谱仪（美国 Wtaers 公司）；ODS 填料（日本 YMC 公司）；D-101 大孔吸附树脂（天津海光化学制药厂）；其余试剂均为分析纯。

甘草药材于 2010 年 7 月采自宁夏灵武市美康甘草基地，经南京中医药大学段金廒教授鉴定为豆科植物乌拉尔甘草 *Glycyrrhiza uralensis* Fisch. 的干燥根及根茎。药材标本（NJUTCM-20100719）存放于南京中医药大学标本馆。

2 提取与分离

称取甘草饮片 10 kg，加入 10 倍量的 50%乙醇加热回流提取 2 h，滤过，药渣重复提取 1 次，滤过，合并提取液并减压浓缩至无醇味；提取物加入适量水配成混悬液，将混悬液采用醋酸乙酯萃取；萃取残余物采用水溶液配制成一定浓度，通过 D-101 大孔树脂洗脱，先以 pH 1～2 纯水洗脱 5 个柱体积，再以 pH 1～2 的 10%乙醇洗脱 5 个柱体积，最后以 pH 1～2 的 50%乙醇洗脱，50%乙醇洗脱液干燥即得甘草总皂苷提取物（0.525 kg）。总皂苷部位（300 g）经 C_{18} 中压柱（900 g）分离，以甲醇-水-乙酸（35∶65∶1→55∶45∶1→75∶25∶1）梯度洗脱，每一梯度溶剂洗脱 5 L，HPLC-PDA-ELSD 跟踪检测，合并相似流分，最终得到 6 个流分 I～VI。I 部位经 C_{18} 中压柱分离，以乙腈-水-乙酸（20∶80∶1→30∶70∶1）梯度洗脱，得到化合物 **1**（22 mg）、**2**（41 mg）、**3**（20 mg）、**5**（25 mg）、**6**（70 mg）。II 部位经 C_{18} 中压柱（900 g）分离，以乙腈-水-乙酸（25∶75∶1→30∶70∶1）梯度洗脱，分离得到化合物 **4**（20 mg）、**7**（50 mg）、**8**（300 mg）、**9**（28 mg）、**10**（27 mg）。III 部位经 C_{18} 中压柱（900 g）分离，以乙腈-水-乙酸（30∶70∶1→40∶60∶1）梯度洗脱，分离得到化合物 **13**（10 g）、**11**（10 mg）、**12**（25 mg）、单葡糖醛酸基甘草次酸（10 mg）。称取化合物 **2**、**6**～**8**、**13** 各 10 mg 置圆底烧瓶中，加入 4 mol/L 盐酸 10 mL，沸水浴加热回流水解 4 h，冷却后用石油醚萃取数次，TLC 检测，浓缩后得皂苷元。

3 结构鉴定

化合物 **1**：白色粉末（甲醇），ESI-MS *m*/*z*: 823 $[M-H]^-$，$C_{42}H_{64}O_{16}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.82 (1H, s, H-12), 5.44 (1H, d, J = 7.0 Hz, H-1″), 5.06 (1H, d, J = 7.0 Hz, H-1′), 3.86 (2H, m, H-30), 3.76 (1H, m, H-22), 3.33 (1H, dd, J = 3.0, 12.0 Hz, H-3), 3.03 (1H, m, H-1), 2.68 (1H, brd, J = 12.0 Hz, H-18), 2.44 (1H, s, H-9), 1.42 (3H, s, H-27), 1.41 (3H, s, H-23), 1.25 (3H, s, H-24), 1.22 (3H, s, H-25), 1.14 (3H, s, H-28), 1.14 (3H, s, H-29), 1.08 (3H, s, H-26), 0.74 (1H, d, J = 12.0 Hz, H-5)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 199.5 (C-11), 172.4 (C-6′), 172.1 (C-6″), 169.4 (C-13), 128.7 (C-12), 107.0 (C-1″), 105.1 (C-1′), 89.2 (C-3), 84.6 (C-2′), 78.5 (C-5″), 77.7 (C-3′), 77.6 (C-3″), 77.5 (C-5′), 76.9 (C-2″), 74.6 (C-22), 73.3 (C-4″), 73.0 (C-4′), 69.9 (C-30), 62.0 (C-9), 55.5 (C-5), 45.4 (C-18), 45.4 (C-8), 44.0 (C-14), 40.6 (C-19), 40.0 (C-4), 39.6 (C-1), 38.6 (C-21), 37.8 (C-17), 37.2 (C-10), 35.9 (C-20), 33.0 (C-7), 28.3 (C-29), 28.1 (C-23), 28.0 (C-16), 26.8 (C-2), 26.6 (C-15), 23.1 (C-27), 21.7 (C-28), 18.9 (C-26), 17.7 (C-6), 16.9 (C-24), 16.8 (C-25)。以上数据与文献报道一致[5]，故鉴定化合物 **1** 为 uralsaponin C。

化合物 **2**：白色粉末（甲醇），ESI-MS *m*/*z*: 849 $[M-H]^-$，$C_{42}H_{57}O_{18}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.77 (1H, s, H-12), 5.44 (1H, d, J = 7.5 Hz, H-1″),

5.05 (1H, d, *J* = 7.5 Hz, H-1′), 4.43 (1H, d, *J* = 5.7 Hz, H-22), 3.36 (1H, dd, *J* = 4.5, 11.7 Hz, H-3), 2.38 (1H, s, H-9), 1.40 (3H, s, H-23), 1.32 (3H, s, H-27), 1.24 (3H, s, H-24), 1.19 (3H, s, H-25), 0.98 (3H, s, H-26), 0.96 (3H, s, H-28), 0.71 (1H, d, *J* = 11.4 Hz, H-5)；^{13}C-NMR (100 MHz, C_5D_5N) *δ*: 198.9 (C-11), 175.1 (C-30), 172.3 (C-6′), 172.1 (C-29), 172.0 (C-6″), 163.9 (C-13), 130.3 (C-12), 106.9 (C-1″), 105.0 (C-1′), 89.1 (C-3), 85.4 (C-22), 84.5 (C-2′), 78.4 (C-5″), 77.7 (C-5′), 77.6 (C-3′), 77.4 (C-3″), 76.8 (C-2″), 73.3 (C-4″), 73.0 (C-4′), 62.0 (C-9), 55.4 (C-5), 53.1 (C-17), 45.1 (C-8), 44.5 (C-18), 44.4 (C-14), 39.9 (C-4), 39.5 (C-1), 37.3 (C-10), 36.2 (C-20), 36.1 (C-21), 35.9 (C-19), 33.1 (C-7), 28.1 (C-23), 26.7 (C-2), 25.9 (C-16), 25.2 (C-15), 23.7 (C-28), 22.2 (C-27), 18.7 (C-26), 17.6 (C-6), 16.9 (C-24), 17.6 (C-25)。以上数据与文献报道一致[5]，故鉴定化合物 **2** 为 uralsaponin D。

化合物 **3**：白色粉末（甲醇），ESI-MS *m*/*z*: 983 $[M-H]^-$，$C_{48}H_{72}O_{21}$。^{1}H-NMR (500 MHz, C_5D_5N) *δ*: 5.92 (1H, s, H-12), 6.34 (1H, d, *J* = 7.6 Hz, H-1‴), 5.41 (1H, d, *J* = 7.5 Hz, H-1′), 5.03 (1H, d, *J* = 7.4 Hz, H-1″), 3.33 (1H, dd, *J* = 4.2, 11.8 Hz, H-3), 2.50 (1H, brd, *J* = 10.5 Hz, H-18), 2.38 (1H, s, H-9), 1.39 (3H, s, H-23), 1.34 (3H, s, H-27), 1.25 (3H, s, H-24), 1.22 (3H, s, H-25), 1.17 (3H, s, H-29), 0.98 (3H, s, H-26), 0.78 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) *δ*: 199.5 (C-11), 175.7 (C-30), 172.0 (C-6″), 172.1 (C-6′), 169.1 (C-13), 128.8 (C-12), 106.9 (C-1″), 105.1 (C-1′), 96.0 (C-1‴), 89.2 (C-3), 84.5 (C-2′), 79.5 (C-3‴), 78.8 (C-5″), 78.4 (C-5‴), 77.6 (C-3′), 77.6 (C-3″), 77.4 (C-5′), 76.8 (C-2″), 74.3 (2‴), 73.3 (C-4″), 73.1 (C-4′), 71.2 (C-4‴), 62.3 (C-6‴), 62.0 (C-9), 55.4 (C-5), 48.2 (C-18), 45.5 (C-14), 44.3 (C-8), 43.4 (C-20), 41.5 (C-19), 39.9 (C-1), 39.5 (C-4), 37.8 (C-22), 37.2 (C-10), 32.9 (C-7), 32.1 (C-17), 31.3 (C-15), 28.5 (C-29), 28.1 (C-23), 27.9 (C-28), 26.8 (C-2), 26.7 (C-16), 23.4 (C-27), 18.7 (C-26), 17.6 (C-6), 16.8 (C-25), 16.7 (C-24)。以上数据与文献报道基本一致[6]，故鉴定化合物 **3** 为 licorice-saponin A3。

化合物 **4**：白色粉末（甲醇-水），ESI-MS *m*/*z*: 893 $[M-H]^-$，$C_{44}H_{64}O_{19}$。^{1}H-NMR (300 MHz, C_5D_5N) *δ*: 5.91 (1H, s, H-12), 5.60 (1H, d, *J* = 7.0 Hz, H-1″), 4.95 (1H, d, *J* = 7.0 Hz, H-1′), 3.40 (1H, dd, *J* = 3.0, 12.0 Hz, H-3), 2.34 (1H, s, H-9), 1.94 (3H, s, $COCH_3$), 1.37 (3H, s, H-23), 1.36 (3H, s, H-27), 1.22 (3H, s, H-25), 1.07 (3H, s, H-29), 0.94 (3H, s, H-26), 0.79 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) *δ*: 199.4 (C-11), 178.9 (C-30), 172.2 (C-6′), 172.3 (C-6″), 170.2 ($COCH_3$), 168.6 (C-13), 128.5 (C-12), 104.5 (C-1″), 104.2 (C-1′), 89.4 (C-3), 80.6 (C-2′), 77.5 (C-5″), 77.4 (C-3′), 77.2 (C-5′), 77.3 (C-3″), 77.1 (C-22), 75.4 (C-2″), 72.8 (C-4′), 72.9 (C-4″), 63.0 (C-24), 61.7 (C-9), 55.6 (C-5), 45.4 (C-18), 44.3 (C-4), 44.1 (C-14), 43.3 (C-8), 40.3 (C-19), 39.7 (C-1), 39.1 (C-20), 36.6 (C-10), 35.7 (C-21), 34.8 (C-17), 32.7 (C-7), 29.1 (C-29), 26.3 (C-2), 26.2 (C-16), 25.4 (C-15), 23.8 (C-27), 22.7 (C-23), 21.5 (C-28), 21.6 (C-28), 20.7 ($COCH_3$), 18.0 (C-6), 18.2 (C-26), 16.3 (C-25)。以上数据与文献报道基本一致[5]，故鉴定化合物 **4** 为 uralsaponin F。

化合物 **5**：白色粉末（甲醇），ESI-MS *m*/*z*: 879 $[M-H]^-$，$C_{44}H_{64}O_{18}$。^{1}H-NMR (300 MHz, C_5D_5N) *δ*: 6.03 (1H, s, H-12), 5.43 (1H, d, *J* = 6.6 Hz, H-1″), 5.05 (1H, d, *J* = 7.6 Hz, H-1′), 3.36 (1H, dd, *J* = 4.4, 11.6 Hz, H-3), 2.46 (1H, s, H-9), 1.47 (3H, s, H-27), 1.42 (3H, s, H-23), 1.33 (3H, s, H-29), 1.24 (3H, s, H-24), 1.21 (3H, s, H-25), 1.08 (3H, s, H-26), 0.93 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) *δ*: 199.5 (C-11), 179.1 (C-30), 172.4 (C-6′), 172.1 (C-6″), 170.2 (-$COCH_3$), 168.3 (C-13), 129.0 (C-12), 106.9 (C-1″), 105.1 (C-1′), 89.2 (C-3), 84.6 (C-2′), 78.4 (C-5″), 77.7 (C-3′), 77.6 (C-3″), 77.4 (C-5′), 77.4 (C-22), 76.8 (C-2″), 73.3 (C-4″), 73.0 (C-4′), 62.2 (C-9), 55.4 (C-5), 45.7 (C-8), 44.6 (C-18), 43.6 (C-14), 40.6 (C-20), 40.1 (C-19), 40.0 (C-4), 39.5 (C-1), 37.2 (C-10), 36.1 (C-17), 35.2 (C-21), 32.7 (C-7), 29.5 (C-29), 28.1 (C-23), 26.7 (C-2), 26.6 (C-15), 25.7 (C-16), 24.1 (C-27), 21.8 (C-28), 20.7 ($COCH_3$), 18.8 (C-26), 17.6 (C-6), 16.9 (C-24), 16.8 (C-25)。以上数据与文献报道基本一致[7]，故鉴定化合物 **5** 为 22β-acetoxyl-glycyrrhizin。

化合物 **6**：白色粉末（甲醇），ESI-MS *m*/*z*: 835 $[M-H]^-$，$C_{42}H_{60}O_{17}$。^{1}H-NMR (300 MHz, C_5D_5N) *δ*: 5.67 (1H, d, *J* = 7.5 Hz, H-1″), 5.00 (1H, d, *J* = 7.5 Hz, H-1′), 3.47 (1H, m, H-3), 2.40 (1H, s, H-9), 1.48

(3H, s, H-23), 1.35 (3H, s, H-27), 1.20 (3H, s, H-25), 1.17 (3H, s, H-29), 0.96 (3H, s, H-26), 0.90 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 199.0 (C-11), 179.6 (C-30), 172.3 (C-6″), 172.1 (C-6′), 164.3 (C-13), 130.0 (C-12), 105.4 (C-1″), 104.7 (C-1′), 89.6 (C-3), 84.0 (C-22), 81.7 (C-2′), 77.9 (C-5′), 77.9 (C-5″), 77.7 (C-3′), 77.7 (C-3″), 75.8 (C-2″), 73.1 (C-4″), 73.1 (C-4′), 63.4 (C-24), 61.9 (C-9), 56.0 (C-5), 45.0 (C-14), 44.9 (C-8), 44.4 (C-4), 44.9 (C-8), 42.1 (C-20), 40.8 (C-19), 39.5 (C-1), 38.1 (C-21), 37.1 (C-10), 35.2 (C-17), 33.5 (C-7), 26.2 (C-2), 25.9 (C-16), 25.2 (C-15), 23.9 (C-28), 23.0 (C-23), 22.3 (C-27), 20.3 (C-26), 18.5 (C-6), 16.7 (C-25)。以上数据与文献报道基本一致[5]，故鉴定化合物 **6** 为 24-hydroxyl-licorice-saponin E2。

化合物 **7**：白色粉末（甲醇），ESI-MS *m*/*z*: 819 $[M-H]^-$，$C_{42}H_{60}O_{16}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.68 (1H, s, H-12), 5.43 (1H, d, *J* = 7.5 Hz, H-1″), 5.04 (1H, d, *J* = 7.5 Hz, H-1′), 3.33 (1H, dd, *J* = 4.2, 11.4 Hz, H-3), 2.40 (1H, s, H-9), 2.28 (1H, dd, *J* = 12.0, 3.5 Hz, H-18), 1.40 (3H, s, H-23), 1.34 (3H, s, H-27), 1.23 (3H, s, H-24), 1.18 (3H, s, H-25), 1.17 (3H, s, H-29), 0.96 (3H, s, H-26), 0.91 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 199.0 (C-11), 179.6 (C-30), 172.3 (C-6″), 172.0 (C-6′), 164.3 (C-13), 130.0 (C-12), 107.0 (C-1″), 105.1 (C-1′), 89.1 (C-3), 84.6 (C-2′), 84.0 (C-22), 78.5 (C-5″), 77.7 (C-5′), 77.6 (C-3′), 77.5 (C-3″), 76.9 (C-2″), 73.3 (C-4″), 73.0 (C-4′), 62.0 (C-9), 55.4 (C-5), 45.0 (C-14), 44.9 (C-8), 44.4 (C-18), 42.1 (C-20), 40.8 (C-19), 39.9 (C-1), 39.5 (C-4), 38.1 (C-21), 37.3 (C-10), 35.7 (C-17), 33.1 (C-7), 28.1 (C-23), 26.7 (C-2), 26.0 (C-16), 25.2 (C-15), 23.9 (C-28), 22.3 (C-27), 20.4 (C-29), 18.7 (C-26), 17.5 (C-6), 16.8 (C-25), 16.7 (C-24)。以上数据与文献报道一致[8]，故鉴定化合物 **7** 为 licorice-saponin E2。

化合物 **8**：白色粉末（甲醇），ESI-MS *m*/*z*: 837 $[M-H]^-$，$C_{42}H_{62}O_{17}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.91 (1H, s, H-12), 5.65 (1H, d, *J* = 7.0 Hz, H-1″), 5.00 (1H, d, *J* = 7.0 Hz, H-1′), 3.46 (1H, dd, *J* = 3.9, 12.6 Hz, H-3), 2.49 (1H, br d, *J* = 11.0 Hz, H-18), 2.41 (1H, s, H-9), 1.46 (3H, s, H-23), 1.40 (3H, s, H-27), 1.31 (3H, s, H-29), 1.18 (3H, s, H-25), 1.00 (3H, s, H-26), 0.74 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 199.3 (C-11), 179.1 (C-30), 172.3 (C-6″), 172.3 (C-6′), 169.6 (C-13), 128.6 (C-12), 105.3 (C-1″), 104.7 (C-1′), 89.7 (C-3), 81.6 (C-2′), 77.9 (C-5″), 77.7 (C-3″), 77.7 (C-5′), 77.4 (C-2″), 75.8 (C-3″, 73.1 (C-4″), 73.1 (C-4′), 63.4 (C-24), 62.0 (C-9), 56.0 (C-5), 48.7 (C-17), 45.5 (C-8), 44.4 (C-4), 44.0 (C-18), 43.4 (C-14), 41.7 (C-19), 39.5 (C-1), 38.4 (C-20), 37.0 (C-10), 33.2 (C-22), 32.1 (C-7), 31.6 (C-21), 28.7 (C-23), 28.6 (C-29), 26.8 (C-28), 26.7 (C-16), 26.6 (C-15), 26.6 (C-2), 23.5 (C-27), 18.5 (C-26), 18.4 (C-6), 16.7 (C-25)。以上数据与文献报道一致[8]，故鉴定化合物 **8** 为 licorice-saponin G2。

化合物 **9**：白色粉末（甲醇-水），ESI-MS *m*/*z*: 863 $[M-H]^-$，$C_{44}H_{64}O_{17}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.88 (1H, s, H-12), 5.43 (1H, d, *J* = 7.0 Hz, H-1″), 5.05 (1H, d, *J* = 7.0 Hz, H-1′), 3.34 (1H, dd, *J* = 3.0, 9.0 Hz, H-3), 2.56 (1H, dd, *J* = 11.5, 3.5 Hz, H-18), 2.41 (1H, s, H-9), 2.03 (3H, s, $COCH_3$), 1.41 (3H, s, H-26), 1.41 (3H, s, H-27), 1.24 (3H, s, H-25), 1.19 (3H, s, H-23), 1.01 (3H, s, H-29), 0.88 (3H, s, H-24), 0.78 (3H, s, H-28), 0.73 (1H, d, *J* = 11.4 Hz, H-5)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 202.8 (C-30), 199.0 (C-11), 172.0 (C-6′), 171.7 (C-6″), 169.5 ($COCH_3$), 166.9 (C-13), 128.8 (C-12), 106.6 (C-1″), 104.8 (C-1′), 88.8 (C-3), 84.3 (C-2′), 78.1 (C-5″), 77.4 (C-5′), 77.3 (C-3′), 77.1 (C-3″), 76.5 (C-2″), 76.3 (C-22), 73.0 (C-4″), 72.7 (C-4′), 61.8 (C-9), 55.0 (C-5), 45.3 (C-8), 44.4 (C-20), 43.4 (C-18), 43.2 (C-14), 39.6 (C-4), 39.1 (C-1), 36.8 (C-10), 36.3 (C-19), 35.5 (C-17), 34.3 (C-21), 32.3 (C-7), 27.8 (C-23), 26.4 (C-2), 26.2 (C-15), 25.2 (C-16), 23.8 (C-27), 23.4 (C-29), 21.1 (C-28), 20.4 ($COCH_3$), 18.3 (C-26), 17.2 (C-6), 16.5 (C-25), 16.4 (C-24)。以上数据与文献报道基本一致[9]，故鉴定化合物 **9** 为 22β-acetoxyl-glyrrhaldehyde。

化合物 **10**：白色粉末（甲醇），ESI-MS *m*/*z*: 807 $[M-H]^-$，$C_{42}H_{64}O_{15}$。^{1}H-NMR (500 MHz, C_5D_5N) δ: 5.72 (1H, s, H-12), 5.42 (1H, d, *J* = 7.5 Hz, H-1″), 5.04 (1H, d, *J* = 7.5 Hz, H-1′), 3.75 (1H, d, *J* = 10.0 Hz, H-30a), 3.69 (1H, d, *J* = 10.0 Hz, H-30b), 3.33 (1H, dd, *J* = 11.6, 4.5 Hz, H-3), 2.40 (1H, s, H-9), 2.22 (1H, dd, *J* = 13.0, 3.5 Hz, H-18), 1.39 (3H, s, H-23), 1.39 (3H, s, H-27), 1.23 (3H, s, H-24), 1.18 (3H, s,

H-25), 1.13 (3H, s, H-29), 1.03 (3H, s, H-26), 0.79 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 199.5 (C-11), 172.4 (C-6′), 172.1 (C-6″), 169.9 (C-13), 128.5 (C-12), 106.9 (C-1″), 105.1 (C-1′), 89.2 (C-3), 84.5 (C-2′), 78.4 (C-5″), 77.7 (C-3′), 77.6 (C-3″), 77.4 (C-5′), 76.8 (C-2″), 36.4 (C-22), 73.2 (C-4″), 73.0 (C-4′), 64.3 (C-30), 62.0 (C-9), 55.4 (C-5), 47.2 (C-18), 45.6 (C-8), 43.5 (C-14), 40.8 (C-19), 39.9 (C-4), 39.5 (C-1), 30.0 (C-21), 32.4 (C-17), 37.2 (C-10), 35.9 (C-20), 32.9 (C-7), 28.2 (C-29), 28.1 (C-23), 26.9 (C-16), 26.7 (C-2), 26.7 (C-15), 23.6 (C-27), 28.7 (C-28), 18.8 (C-26), 17.6 (C-6), 16.8 (C-24), 16.6 (C-25)。以上数据与文献报道基本一致[7]，故鉴定化合物 **10** 为 3β-*O*-[β-*D*-glucuronopyranosyl-(1→2)-β-*D*-glucuronopyranosyl]-glycyrretol。

化合物 **11**：白色粉末（甲醇-水），ESI-MS *m*/*z*: 777 $[M-H]^-$，$C_{41}H_{62}O_{14}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.96 (1H, s, H-12), 5.43 (1H, d, *J* = 7.6 Hz, H-1″), 5.01 (1H, d, *J* = 7.6 Hz, H-1′), 3.41 (1H, dd, *J* = 4.6, 11.6 Hz, H-3), 2.41 (1H, s, H-9), 1.45 (3H, s, H-23), 1.33 (3H, s, H-27), 1.32 (3H, s, H-29), 1.25 (3H, s, H-24), 1.13 (3H, s, H-25), 1.09 (3H, s, H-26), 0.79 (3H, s, H-28)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 199.4 (C-11), 179.1 (C-30), 172.3 (C-6′), 168.9 (C-13), 128.6 (C-12), 106. 7 (C-1″), 105.2 (C-1′), 88.9 (C-3), 83.4 (C-2′), 66.6 (C-5″), 77.4 (C-3′), 74.1 (C-3″), 77.4 (C-5′), 38.6 (C-22), 73.5 (C-2″), 69.1 (C-4″), 73.0 (C-4′), 62.2 (C-9), 55.6 (C-5), 48.6 (C-18), 45.7 (C-8), 43.8 (C-14), 44.3 (C-20), 42.1 (C-19), 40.1 (C-4), 39.6 (C-1), 37.4 (C-10), 32.4 (C-17), 31.7 (C-21), 33.3 (C-7), 29.0 (C-29), 28.2 (C-23), 26.9 (C-2), 26.8 (C-15), 26.7 (C-16), 24.0 (C-27), 28.8 (C-28), 19.4 (C-26), 18.1 (C-6), 17.1 (C-24), 17.0 (C-25)。以上数据与文献报道基本一致[10]，故鉴定化合物 **11** 为 araboglycyrrhizin。

化合物 **12**：白色粉末（甲醇），ESI-MS *m*/*z*: 823 $[M-H]^-$，$C_{42}H_{64}O_{16}$。^{1}H-NMR (300 MHz, C_5D_5N) δ: 5.73 (1H, d, *J* = 7.4 Hz, H-1′), 5.45 (1H, brs, H-12), 5.03 (1H, d, *J* = 7.4 Hz, H-1″), 4.39 (1H, d, *J* = 10.5 Hz, H-24a), 3.64 (1H, d, *J* = 10.0 Hz, H-24b), 3.46 (1H, dd, *J* = 4.2, 12.4 Hz, H-3), 2.42 (1H, dd, *J* = 13.3, 3.5 Hz, H-18), 1.65 (3H, s, H-27), 1.48 (3H, s, H-23), 1.37 (3H, s, H-29), 0.90 (3H, s, H-26), 0.87 (3H, s, H-28), 0.78 (3H, s, H-25)；^{13}C-NMR (100 MHz, C_5D_5N) δ: 179.6 (C-30), 172.3 (C-6′), 172.0 (C-6″), 145.1 (C-13), 122.8 (C-12), 105.2 (C-1″), 104.7 (C-1′), 89.9 (C-3), 81.3 (C-2′), 77.9 (C-5″), 77.7 (C-3′), 77.7 (C-3″), 77.7 (C-5′), 75.7 (C-2″), 73.1 (C-4″), 73.0 (C-4′), 63.4 (C-24), 56.2 (C-5), 48.6 (C-17), 47.7 (C-9), 44.3 (C-18), 44.0 (C-8), 43.5 (C-14), 41.7 (C-19), 40.0 (C-20), 39.1 (C-1), 38.7 (C-4), 36.6 (C-10), 33.2 (C-22), 32.3 (C-7), 31.8 (C-21), 29.1 (C-29), 28.5 (C-28), 27.3 (C-16), 26.6 (C-15), 26.6 (C-2), 26.1 (C-27), 23.9 (C-11), 22.9 (C-23), 19.1 (C-6), 16.8 (C-26), 15.5 (C-25)。以上数据与文献报道基本一致[7]，故鉴定化合物 **12** 为 licorice-saponin J2。

化合物 **13**：白色粉末（MeOH），与甘草酸对照品共薄层，Rf 及显色行为均一致，液相检测保留时间与最大吸收波长均一致，与甘草酸对照品混合熔点不下降，故鉴定化合物 **13** 为甘草酸。

化合物 **14**：白色粉末（MeOH），ESI-MS *m*/*z*: 644 $[M-H]^-$，$C_{36}H_{54}O_{10}$。^{1}H-NMR (500 MHz, C_5D_5N) δ: 5.97 (1H, brs, H-12), 5.73 (1H, d, *J* = 7.6 Hz, H-1′), 3.45 (1H, dd, *J* = 4.5, 12.5 Hz, H-3), 2.54 (1H, dd, *J* = 12.5, 3.0 Hz, H-18), 2.50 (1H, s, H-9), 1.45 (3H, s, H-27), 1.35 (3H, s, H-23), 1.35 (3H, s, H-29), 1.25 (3H, s, H-25), 1.09 (3H, s, H-24), 1.04 (3H, s, H-26), 0.80 (3H, s, H-28)；^{13}C-NMR (125 MHz, C_5D_5N) δ: 39.5 (C-1), 26.7 (C-2), 88.8 (C-3), 39.9 (C-4), 55.4 (C-5), 17.7 (C-6), 33.0 (C-7), 45.6 (C-8), 62.1 (C-9), 37.3 (C-10), 199.5 (C-11), 128.7 (C-12), 169.6 (C-13), 43.5 (C-14), 26.9 (C-15), 26.7 (C-16), 32.2 (C-17), 48.7 (C-18), 41.7 (C-19), 44.1 (C-20), 31.6 (C-21), 38.4 (C-22), 28.2 (C-23), 16.8 (C-24), 17.0 (C-25), 18.8 (C-26), 23.6 (C-27), 28.7 (C-28), 28.8 (C-29), 179.1 (C-30), 107.2 (C-1′), 78.2 (C-2′), 75.5 (C-3′), 73.4 (C-4′), 77.9 (C-5′), 172.8 (C-6′)。以上数据与文献报道基本一致[11]，故鉴定化合物 **14** 为单葡糖醛酸基甘草次酸。

4 生物活性及讨论

采用 CCK-8 检测法[12]，测定化合物 **1**～**14** 及 **2**、**6**～**8**、**13** 水解后的苷元对 3 种人源肿瘤细胞株 MGC-803、SW620、SMMC-7721 体外增殖的影响，结果发现皂苷化合物均无显著的细胞毒活性（IC_{50}＞100 μmol/L，水解后苷元细胞毒活性增强（表 1）。

表 1 化合物 2、6～8、13 的水解后苷元对 3 种人源肿瘤的细胞毒活性

Table 1 Cytotoxic activity of aglycones of compounds 2, 6—8, and 13 after hydrolysis on three kinds of human cancer cell lines

化合物	IC_{50} / (μmol·L^{-1})		
	MGC-803	SW620	SMMC-7721
2 的苷元	25.6±3.5	26.5±3.2	29.1±2.1
6 的苷元	41.6±1.4	34.9±1.6	38.4±2.7
7 的苷元	31.3±1.4	30.4±3.1	27.3±1.5
8 的苷元	36.9±2.5	31.2±2.3	36.5±2.1
13 的苷元	20.5±1.6	21.9±1.0	18.3±0.8

Kanaoka 等[11]在研究甘草酸的代谢产物时，从人的血清中分离得到单葡糖醛酸基甘草次酸。Glavac 等[13]发现口服甘草酸后，在人的尿液中也检测到化合物 **14**。此外，发现人源肠内生菌也能将甘草酸转化成单葡糖醛酸基甘草次酸[14]。药理研究表明，该化合物具有多种药理作用[15]。本实验是首次报道从天然产物中分离得到单葡糖醛酸基甘草次酸，并且采用 UPLC-MS 从甘草的 50%甲醇提取液中检测到该化合物，这不仅提示天然存在单葡糖醛酸基甘草次酸，也为揭示甘草的药理作用提供科学依据。

参考文献

[1] Fiore C, Eisenhut M, Krausse R, *et al*. Antiviral effects of *Glycyrrhiza* species [J]. *Phytother Res*, 2008, 22(2): 141-148.

[2] 罗祖良, 李 倩, 覃洁萍, 等. 光果甘草的研究进展 [J]. 中草药, 2011, 42(10): 2154-2158.

[3] Ashfaq U A, Masoud M S, Nawaz Z, *et al*. Glycyrrhizin as antiviral agent against hepatitis C virus [J]. *J Transl Med*, 2011, 9: 112.

[4] Zhang Q Y, Ye M. Chemical analysis of the Chinese herbal medicine Gan-Cao (licorice) [J]. *J Chromatogr A*, 2009, 1216(11): 1954-1969.

[5] Zheng Y F, Qi L W, Cui X B, *et al*. Oleanane-type triterpene glucuronides from the roots of *Glycyrrhiza uralensis* Fischer [J]. *Planta Med*, 2010, 76(13): 1457-1463.

[6] Kitagawa I, Hori K, Taniyama T, *et al*. Saponin and sapogenol. 47. on the constituents of the roots of *Glycyrrhiza-uralensis* Fischer from Northeastern China. 1. Licorice-Saponins A3, B2, and C2 [J]. *Chem Pharm Bull*, 1993, 41(1): 43-49.

[7] Yang J W, Nakajima J, Kimura N, *et al*. Oleanane-type triterpene glycosides from *Glycyrrhiza uralensis* [J]. *Nat Prod Commun*, 2007, 2(3): 243-248.

[8] Kitagawa I, Hori K, Sakagami M, *et al*. Saponin and sapogenol. 48. on the constituents of the roots of *Glycyrrhiza-uralensis* Fischer from Northeastern China. 2. Licorice-saponins D3, E2, F3, G2, H2, J2, and K2 [J]. *Chem Pharm Bull*, 1993, 41(8): 1337-1345.

[9] 张 慧, 王世盛, 李 伟, 等. 乌拉尔甘草中的一个新三萜皂苷成分 [J]. 世界科学技术—中医药现代化, 2009, 11(2): 253-256.

[10] Kitagawa I, Hori K, Sakagami M, *et al*. Saponin and sapogenol. 49. on the constituents of the roots of *Glycyrrhiza-inflata* Batalin from Xinjiang, China-characterization of 2 sweet oleanane-type triterpene oligoglycosides, spioglycyrrhizin and sraboglycyrrhizin [J]. *Chem Pharm Bull*, 1993, 41(8): 1350-1357.

[11] Kanaoka M, Yano S, Kato H, *et al*. Synthesis and separation of 18β-glycyrrhetyl monoglucuronide from serum of a patient with glycyrrhizin-induced pseudo-aldosteronism [J]. *Chem Pharm Bull*, 1986, 34(12): 4978-4983.

[12] 朱登祥, 安 芳, 王书华, 等. 牡荆苷对人食管癌 EC-109 细胞生长及凋亡的影响 [J]. 中草药, 2012, 43(9): 1781-1784.

[13] Glavac N K, Kreft S. Excretion profile of glycyrrhizin metabolite in human urine [J]. *Food Chem*, 2012, 131(1): 305-308.

[14] Akao T. Effect of pH on metabolism of glycyrrhizin, glycyrrhetic acid and glycyrrhetic acid monoglucuronide by collected human intestinal flora [J]. *Chem Pharm Bull*, 2001, 24(10): 1108-1112.

[15] Mizutani K, Kambara T, Masuda H, *et al*. Glycyrrhetic acid monoglucuronide (MGGR): biological activities [A] // *Towards Natural Medicine Research in the 21st Century* [C]. Amsterdam: ELsevier Science, 1998.