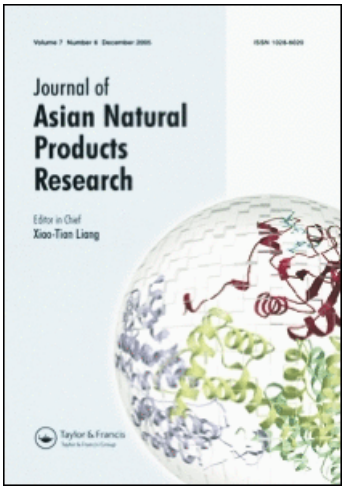


Journal of Asian Natural Products Research
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713454007

Three new xanthones from the roots of *Polygala japonica* Houtt.

Qing-Chun Xue[a]; Chuang-Jun Li[a]; Li Zuo[a]; Jing-Zhi Yang[a]; Dong-Ming Zhang[a]
[a] Key Laboratory of Bioactive Substances and Resources Utilization of Chinese Herbal Medicine, Ministry of Education, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

To cite this Article Xue, Qing-Chun , Li, Chuang-Jun , Zuo, Li , Yang, Jing-Zhi and Zhang, Dong-Ming(2009) 'Three new xanthones from the roots of *Polygala japonica* Houtt.', Journal of Asian Natural Products Research, 11: 5, 465 — 469
To link to this Article: DOI: 10.1080/10286020902835547
URL: http://dx.doi.org/10.1080/10286020902835547

Journal of Asian Natural Products Research
Vol. 11, No. 5, May 2009, 465–469

Taylor & Francis
Taylor & Francis Group

Three new xanthones from the roots of *Polygala japonica* Houtt.

Qing-Chun Xue, Chuang-Jun Li, Li Zuo, Jing-Zhi Yang and Dong-Ming Zhang*

Key Laboratory of Bioactive Substances and Resources Utilization of Chinese Herbal Medicine, Ministry of Education, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100050, China

(*Received 6 January 2009; final version received 19 February 2009*)

Three new xanthones, 1,3-dihydroxy-2,5,6,7-tetramethoxyxanthone (**1**), 3-hydroxy-1,2,5,6,7-pentamethoxyxanthone (**2**), and 3,8-dihydroxy-1,2,6-trimethoxyxanthone (**3**), together with two known compounds were isolated from the roots of *Polygala japonica* Houtt. Their structures were determined on the basis of spectroscopic data.

Keywords: *Polygala japonica* Houtt.; 1,3-dihydroxy-2,5,6,7-tetramethoxyxanthone; 3-hydroxy-1,2,5,6,7-pentamethoxyxanthone; 3,8-dihydroxy-1,2,6-trimethoxyxanthone

1. Introduction

Polygala japonica Houtt., belonging to the genus *Polygala* of the family Polygalaceae, is a folk medicinal plant in the south of China and has been used to treat pharyngolaryngitis, tonsillitis, stomatitis, pneumonia, calculus, wound, and diphtheria for a long time. Our laboratory has done research both on the MeOH extract and EtOAc extract in which a series of triterpenoid saponins and xanthones had been discovered [1]. In this report, we investigated the $CHCl_3$ extract of this plant, which resulted in the isolation of three new xanthones, named 1,3-dihydroxy-2,5,6,7-tetramethoxyxanthone (**1**), 3-hydroxy-1,2,5,6,7-pentamethoxyxanthone (**2**), and 3,8-dihydroxy-1,2,6-trimethoxyxanthone (**3**) as well as two known compounds 1,7-dihydroxy-2,3,4-trimethoxyxanthone (**4**) and 1,7-dihydroxy-3,4-dimethoxyxanthone (**5**).

2. Results and discussion

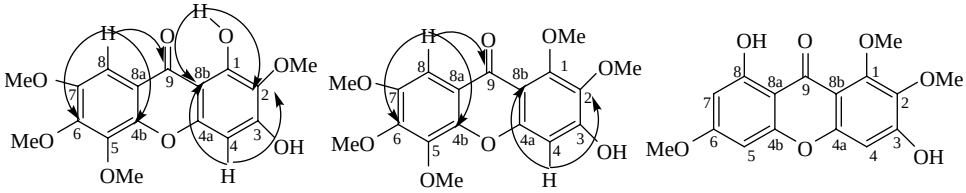
The $CHCl_3$ soluble parts of the 95% EtOH extract of *P. japonica* were subjected to silica-gel column chromatography repeatedly and purified by preparative HPLC to afford three new xanthones and two known compounds. On the basis of the spectral data and by comparing their spectral data with those of reported in the literature, the structures of these xanthones were determined as follows.

Compound **1** was obtained as yellow powder, whose molecular formula was established as $C_{17}H_{16}O_8$ by HR-EI-MS at *m/z* 348.0856 $[M]^+$ and 347.0784 $[M-H]^-$. The xanthone skeleton was suggested by the UV absorption maxima at 204, 239, 260, 319, and 367 nm and the ^{13}C NMR spectra which showed 17 carbon signals including one carbonyl group (δ 180.5), 12 aromatic carbons, and 4 methoxyl carbons. The ^1H NMR spectrum showed four methoxyl groups at δ 3.96 (s, 3H), 4.04 (s, 6H), and 4.07 (s, 3H), two singlets of aromatic protons at δ 6.61 and 7.40, assigned to H-4 and H-8, respectively [2], and a singlet of hydroxyl

*Corresponding author. Email: zhangdm@imm.ac.cn

ISSN 1028-6020 print/ISSN 1477-2213 online

DOI: 10.1080/10286020902835547
http://www.informaworld.com



1 **2** **3**

Figure 1. The structure of compounds **1**–**3**.

proton at δ 13.13, which indicated the existence of intramolecular hydrogen bond. In the HMBC spectrum (Figure 1), the aromatic proton at δ 6.61 (H-4) correlated with the carbons at δ 106.6 (C-8b), 129.6 (C-2), 153.2 (C-4a), 156.1 (C-3). As a result, it was confirmed at position 4. The other aromatic proton at δ 7.40 (H-8) correlated with the carbons at δ 115.5 (C-8a), 145.9 (C-4b), 148.6 (C-6), 150.1 (C-7), 180.5 (C-9), so we could easily confirm its position 8 [2]. The above conclusion was further confirmed by the NOE experiment in which the aromatic proton at δ 7.40 (H-8) was correlated only with the methoxyl group at δ 3.96, meanwhile, the aromatic proton at δ 6.61 (H-4) was not correlated with any other group. Further structure elucidation of this compound was made by the comparison of its NMR spectral data with the known compound 1,3,8-trihydroxy-2,6-dimethoxyxanthone [3]. Thus, compound **1** was elucidated as 1,3-dihydroxy-2,5,6,7-tetramethoxyxanthone (Figure 1). The structure of **1** was confirmed and all signals were assigned by the aid of the 2D NMR spectra (Figure 1; Table 1).

Compound **2** was obtained as yellow powder, whose molecular formula was established as $C_{18}H_{18}O_8$ by HR-EI-MS at *m/z* 362.1015. The UV spectrum also suggested the presence of a xanthone skeleton by showing the absorption maxima at 208, 241, 250, 312, and 352 nm and the skeleton was confirmed by the ^{13}C NMR spectra showing 18 carbon signals including one carbonyl group (δ 174.7), 12 aromatic carbons, and 5 methoxyl

Table 1. ^{13}C NMR spectral data of compounds **1** (125 MHz), **2** (100 MHz), and **3** (125 MHz).

No.	Compound **1**[a]	Compound **2**[a]	Compound **3**[b]
1	153.1	152.3	161.2
2	129.6	137.7	139.7
3	156.1	155.1	152.9
4	93.5	99.3	96.6
4a	153.2	154.5	154.1
4b	145.9	144.7	156.2
5	141.3	141.1	91.7
6	148.6	147.6	162.7
7	150.1	149.9	99.6
8	100.1	101.0	165.1
8a	115.5	117.7	105.4
8b	106.6	110.0	102.6
9	180.5	174.7	178.9
OCH_3	56.3	61.9	55.8
OCH_3	60.9	61.8	60.5
OCH_3	61.5	61.7	61.5
OCH_3	62.0	61.4	
OCH_3		59.8	

[a] Measured in $CDCl_3$.
[b] Measured in DMSO-d_6.

carbons. The ^{1}H NMR spectra showed five methoxyl groups at δ 4.03 (s, 6H), 4.01 (s, 3H), 4.00 (s, 3H), and 3.94 (s, 3H) and two aromatic protons at δ 6.87 and 7.47 [2]. The NMR spectral data indicated the structure of compound **2** was almost identical to that of compound **1**, except that the methoxyl at C-1 position substituted the hydroxyl of compound **1**. Thus, compound **2** was elucidated as 3-dihydroxy-1,2,5,6,7-pentamethoxyxanthone (Figure 1).

Compound **3** was isolated as a yellow powder. Based on the HR-EI-MS of **3** indicating $[M]^+$ ion at *m/z* 318.0742, its molecular formula was deduced to be $C_{16}H_{14}O_7$. The presence of a xanthone skeleton was suggested by the UV spectrum showing absorption maxima at 205, 241, 312, and 347 nm and the ^{13}C NMR spectrum (Table 1) showing 16 carbon signals including one carbonyl group (δ 178.9), 12 aromatic carbons, and 3 methoxyl carbons. The ^{1}H NMR spectrum of **3** showed the presence of three methoxyl signals at δ 3.77, 3.81, and 3.82, three aromatic protons at δ 6.23 (1H, s), 6.43 (1H, d, $J = 2.0$ Hz), and 6.58 (1H, d, $J = 2.0$ Hz), and a singlet of hydroxyl proton at δ 13.69, which indicated that the hydroxyl should be located at C-1 or C-8 position. In addition, the NOE experiment showed that there were cross-correlation peaks between both the aromatic protons at δ 6.43 and 6.58 and the methoxyl group at δ 3.77, from which, we could come to the conclusion that the aromatic protons at δ 6.43 and 6.58 were on the *meta*-position, respectively [4]. Besides, the NOE experiment also showed that there were no cross-correlation peaks between the aromatic protons at δ 6.23 and any other group indicated that the aromatic proton at δ 6.23 was at the C-4 position [4] and one hydroxyl was attributed to the position C-3. As a result, by comparing with the known compound 1,8-dihydroxy-2,3,4,6-tetramethoxyxanthone [4] and the compound **2**, **3** was elucidated as 3,8-dihydroxy-1,2,6-trimethoxyxanthone (Figure 1) and all the signals were assigned by comparing with 1,8-dihydroxy-2,3,4,6-tetramethoxyxanthone [4] and compound **2**.

3. Experimental

3.1 General experimental procedures

UV spectra were measured on a JASCO V-650 spectrophotometer. IR spectra were measured on a Nicolet 5700 spectrometer as KBr pellets. NMR spectra were run on a Varian Inoval-400, 500 Spectrometer using TMS as the internal standard. ESI-MS were measured on an Aglilent 1100 series LC/MSD Trap SL mass spectrometer and an Autospec-Ultima ETOF, respectively. Silica-gel (100–200, 200–300 mesh; Qingdao Marine Chemical Inc., Qingdao, China) and silica-gel GF-254 (Qingdao Marine Chemical Inc.) were used for column chromatography and TLC, respectively.

3.2 Plant material

The roots of *P. japonica* were collected on June 2003 in Jiangxi Province, China, which was identified by Professor Yongming Luo, Jiangxi College of Traditional Chinese Medicine. A voucher specimen (No. 272400) is deposited in Institute of Materia Medica, Chinese Academy of Medical Science and Peking Union Medical College, Beijing.

3.3 Extraction and isolation

Dried roots of *P. japonica* (5 kg) were extracted three times with 95% EtOH for 2 h each time, obtained 0.839 kg residue after removal of the solvent under reduced pressure. This residue was subjected to silica-gel column chromatography (100–200 mesh), eluted with $CHCl_3$, EtOAc, EtOAc:acetone (1:1), EtOAc:acetone (1:3), acetone, acetone:EtOH (1:1), EtOH and MeOH, successively. The $CHCl_3$ extract (40 g) was subjected to silica-gel column chromatography (100–200 mesh), eluting first with $CHCl_3$, then with a gradient of $CHCl_3$–MeOH (100/1 → 100/5) to yield

26 fractions. Fractions 10 and 11 were separated by silica-gel column chromatography (200–300 mesh), eluting first with a gradient of EtOAc–petroleum ether (1/15 → 1/4) to yield 300 fractions. From fractions 190 to 250, by preparative HPLC [ODS C-18; detective wavelength: 254 nm; flow rate: 7 ml/min; mobile phase: CH_3OH/H_2O (60:40)], compound **1** (6 mg, retain time: 70 min) was obtained. Fraction 17 was separated by silica-gel column chromatography (200–300 mesh), eluting first with a gradient of acetone–petroleum ether (2/7 → 1/2) to yield 60 fractions. From fractions 23 to 26, by preparative HPLC [ODS C-18; detective wavelength: 254 nm; flow rate: 7 ml/min; mobile phase: CH_3OH/H_2O (60:40)], compound **3** (30 mg, retain time: 98 min) was obtained. Fraction 19 was separated by silica-gel column chromatography (200–300 mesh), eluting first with a gradient of acetone–petroleum ether (1/6 → 1/3) to yield 300 fractions. From fractions 250 to 300, by preparative HPLC [ODS C-18; detective wavelength: 254 nm; flow rate: 7 ml/min; mobile phase: CH_3OH/H_2O (53:47)], compound **2** (15 mg, retain time: 50 min) was obtained.

*3.3.1 1,3-Dihydroxy-2,5,6,7-tetramethoxyxanthone (**1**)*

Yellow powder; UV (MeOH) λ_{max} (nm) ($\log \varepsilon$): 204 (4.34), 239 (4.08), 260 (4.10), 319 (3.89), 367 (3.48); IR (KBr) ν_{max} (cm^{-1}): 3119, 2948, 1640, 1567, 1478; ^1H NMR ($CDCl_3$, 400 MHz) δ (ppm) 13.13 (1H, s, OH-1), 7.40 (1H, s, H-8), 6.61 (1H, s, H-4), 4.07 (3H, s, OCH_3-2/5/6), 4.04 (6H, s, OCH_3-2/5/6), 3.96 (3H, s, OCH_3-7). For ^{13}C NMR spectral data, see Table 1; ESI-MS *m/z*: 347.1 $[M-H]^-$; HR-EI-MS *m/z*: 348.0856 $[M]^+$ (calcd for $C_{17}H_{16}O_8$, 348.0845).

*3.3.2 3-Hydroxy-1,2,5,6,7-pentamethoxyxanthone (**2**)*

Yellow powder; UV (MeOH) λ_{max} (nm) ($\log \varepsilon$): 208 (3.81), 241 (3.90), 250 (3.89), 312 (3.67), 352 (3.34); IR (KBr) ν_{max} (cm^{-1}): 3097, 2993, 2943, 2842, 1573, 1468; ^1H NMR ($CDCl_3$, 500 MHz) δ (ppm) 7.47 (1H, s, H-8), 6.87 (1H, s, H-4), 4.03 (6H, s, OCH_3-5/6), 4.01 (3H, s, OCH_3-1/2), 4.00 (3H, s, OCH_3-1/2), 3.94 (3H, s, OCH_3-7). For ^{13}C NMR spectral data, see Table 1; ESI-MS *m/z*: 361.2 $[M-H]^-$; HR-EI-MS *m/z*: 362.1015 $[M]^+$ (calcd for $C_{18}H_{18}O_8$, 362.1002).

*3.3.3 3,8-Dihydroxy-1,2,6-trimethoxyxanthone (**3**)*

Yellow powder; UV (MeOH) λ_{max} (nm) ($\log \varepsilon$): 205 (4.44), 241 (4.39), 312 (4.20), 347 (3.85); IR (KBr) ν_{max} (cm^{-1}): 3253, 1665, 1591, 1508, 1477; ^1H NMR (DMSO-d_6, 500 MHz) δ (ppm) 13.69 (1H, s, OH-8), 6.58 (1H, s, H-4), 6.43 (1H, d, $J = 2.0$ Hz, H-5/H-7), 6.23 (1H, d, $J = 2.0$ Hz, H-5/H-7), 3.82 (3H, s, OCH_3-1/2), 3.81 (3H, s, OCH_3-1/2), 3.77 (3H, s, OCH_3-6). For ^{13}C NMR spectral data, see Table 1; ESI-MS *m/z*: 317.0 $[M-H]^-$; HR-EI-MS *m/z*: 318.0742 $[M]^+$ (calcd for $C_{16}H_{14}O_7$, 318.0739).

The known compounds were characterized as 1,7-dihydroxy-2,3,4-trimethoxy-xanthone and 1,7-dihydroxy-3,4-dimethoxy-xanthone, whose data were identical to those reported in the literature [5].

Acknowledgements

Financial support for this research was provided by the National Science Foundation of China (Nos 20372087 and 204320307). We are grateful to the Department of Instrumental Analysis, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College for all spectra measurements.

References

[1] J. Fu, D.M. Zhang, and R.Y. Chen, *J. Asian Nat. Prod. Res.* **8**, 41 (2006).

[2] A. Kijjoa, M.J. Gonzales, C.M. Afonso, M.M.M. Pinto, C. Anantachoke, and W. Herz, *Phytochemistry* **53**, 1021 (2000).
[3] E.P. Ortega, R.E. Lopez-Garcia, R.M. Rabanal, V. Darias, and S. Valverde, *Phytochemistry* **27**, 1912 (1988).
[4] W.G. Van, D. Sluis, and R.P. Labadie, *Phytochemistry* **24**, 2601 (1985).
[5] A. Bashir, M. Hamburger, J.D. Msonthi, and K. Hostettmann, *Phytochemistry* **31**, 309 (1992).