Fitoterapia 125 (2018) 184–190

Contents lists available at ScienceDirect

Fitoterapia

journal homepage: www.elsevier.com/locate/fitote

Flavonoid glycosides and alkaloids from the embryos of *Nelumbo nucifera* seeds and their antioxidant activity

Xi-Lan Jiang[a,b,c], Lun Wang[a,b,*], En-Jun Wang[b], Guo-Lin Zhang[b], Bin Chen[b], Ming-Kui Wang[a,b], Fu Li[a,b,*]

[a] School of Pharmacy and Bioengineering, Chengdu University, Chengdu 610106, China
[b] Key Laboratory of Mountain Ecological Restoration and Bioresource Utilization and Ecological Restoration, Biodiversity Conservation Key Laboratory of Sichuan Province, Chengdu Institute of Biology, Chinese Academy of Sciences, Chengdu 610041, China
[c] Key Laboratory of Exploitation and Study of Distinctive Plants in Education Department of Sichuan Province, Sichuan University of Arts and Science, Dazhou 635000, China

ARTICLE INFO

Keywords:
Nelumbo nucifera
Flavonoid glycosides
Alkaloids
Antioxidant activity

ABSTRACT

Chemical investigation of the embryos of *Nelumbo nucifera* afforded four new flavone C-glycosides, named nelumbosides A–D (**1–4**), together with nine known ones, comprising five flavonoids (**5–9**) and four alkaloids (**10 – 13**). The chemical structures of the new compounds were elucidated by 1D, 2D-NMR and HR-ESI-MS techniques, together with chemical methods. Nelumbosides A–D (**1–4**) are rarely present in naturally occurring flavone C-glycosides featuring a 4-hydroxystyrene unit connected to the flavonoid skeleton. Compounds **2–13** were evaluated for their antioxidant activity by ABTS and DPPH radical-scavenging assay. Among them, compounds **2**, **6**, **7** and **11** exhibited strong scavenging activity with SC_{50} values ranging from 12.07 to 25.68 μM compared with the positive control L-ascorbic acid.

1. Introduction

Nelumbo nucifera GAERTNER, considered to be an economically important perennial aquatic crop, has been cultivated all over China [1]. Many parts of this plant, such as leaves, flowers, rhizomes and embryos, have been used as common vegetable, tea and healthcare product [2]. Meanwhile, the different organs of this plant have also been used in traditional herbal medicine since ancient times to cure different diseases [3]. The embryos of *N. nucifera* seeds, also called "Lian Zi Xin", were primarily used to remove heat from the heart, anchor the mind, improve spermatorrhea and arrest bleeding [4]. Modern pharmaceutical studies showed that alkaloids from the embryos of *Nelumbo nucifera* seeds possessed various biological activities, including anti-tumor [5–6], anti-inflammatory [7], anti-oxidation [8–9] and obvious sedation activities [10–11].

Previous phytochemical studies on the embryos of *N. nucifera* seeds indicated the presence of alkaloids [11–13], sterols and fatty alcohols [14]. A recent study reported the profiling of flavonoids in Lotus (*Nelumbo nucifera*) by HPLC-DAD and HPLC-ESI-MS^n [15]. However, detailed phytochemical study on the flavonoids in this plant has rarely been reported. Moreover, flavonoids and alkaloids in this plant were considered to be the bioactive components. The lack of chemical entities greatly limited biological evaluation of this important resource. Therefore, the present study focused on the investigation of these two kinds of compounds in the embryos of *N. nucifera* seeds in order to provide a scientific foundation for further use of this plant. As a result, four novel flavone C-glycosides (**1–4**), with an unusual structure featuring a 4-hydroxystyrene unit connected to the flavonoid skeleton, together with nine known compounds (**5–13**) were isolated from the embryos of *N. nucifera* seeds (Fig. 1). Herein, the isolation and identification of these compounds and their antioxidant activities against $ABTS^{+\cdot}$ and DPPH assay are reported.

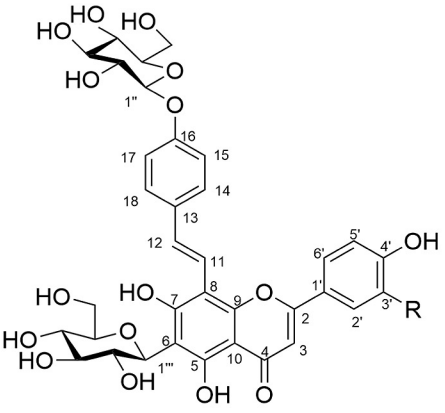
2. Experimental

2.1. General experimental procedures

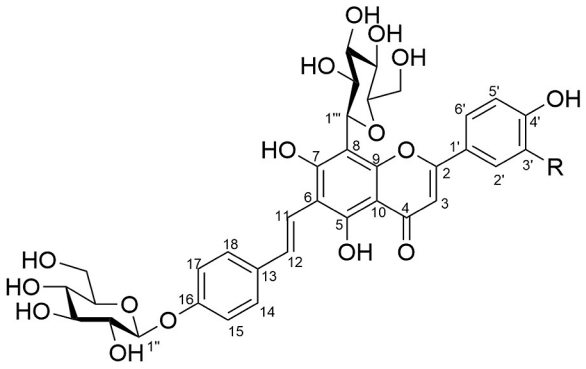
Optical rotations were determined on a Perkin-Elmer 341 polarimeter at room temperature. IR spectra were carried out on a Perkin-Elmer 1725X-FT spectrometer with KBr disks. UV spectra were taken in MeOH on a Shimadzu UV 260 Spectrometer. ^{1}H NMR and ^{13}C NMR, HSQC and HMBC spectra were recorded with a Bruker Avance-600 spectrometer and a Bruker Avance-400 spectrometer (Bruker Corporation, Stockholm, Sweden). The chemical shift (δ) values are

* Corresponding authors at: School of Pharmacy and Bioengineering, Chengdu University, Chengdu 610106, China.
E-mail addresses: wanglun@cib.ac.cn (L. Wang), lifu@cib.ac.cn (F. Li).

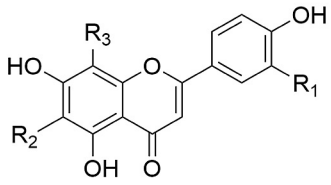
https://doi.org/10.1016/j.fitote.2018.01.009
Received 5 December 2017; Received in revised form 17 January 2018; Accepted 21 January 2018
Available online 31 January 2018



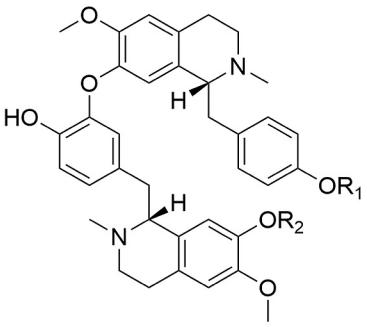
1 R=OH
3 R=H



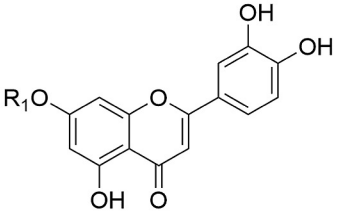
2 R=OH
4 R=H



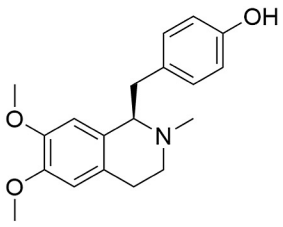
	R_1	R_2	R_3
5	H	β - D - glucopyranoside	β - D - xylopyranosyl
6	OH	H	β - D - glucopyranoside
7	OH	β - D - glucopyranoside	H
8	H	β - D - glycopyranoside	β - D - arabinosyl



	R_1	R_2
10	CH_3	CH_3
11	CH_3	H
12	H	CH_3



9 α - L - rhamnopyranosyl-（1→2）- β - D - glucopyranoside



13

Fig. 1. Structures of compounds **1–13**.

given in ppm with TMS as internal standard, and coupling constants (*J*) are in Hz. ESI-MS and HR-ESI-MS were measured on a Finnigan LCQ^{DECA} (San Jose, CA, USA) and a Bruker Apex-III mass spectrometer (Bruker Corporation, Stockholm, Sweden) respectively. Preparative HPLC was performed using Ultimate ® XB-C_{18} (250 mm × 21.2 mm, 10 μm) preparative column. ABTS and DPPH free radical-scavenging activities were measured by using Thermo Scientific Microplate Reader (Thermo Fisher Scientific). Sephadex LH-20 (GE); RP-C_{18} (Welch); silica gel (100–200 mesh and 200–300 mesh, Qingdao Marine Chemical Factory) and silica gel 60 (40–63 μm, Merck, Germany) were used for column chromatography (CC). TLC was carried out on precoated silica gel GF_{254} (10–40 μm, Qingdao Marine Chemical Factory) plates; Spots were visualized under UV light and by spraying with 10% H_2SO_4 in C_2H_5OH (v/v) followed by heating. The solvents used for extraction and isolation were analytical grade (Chengdu Kelong Chemical Reagent Factory, Chengdu, China). The ratios of solvent are described as a mixture by v/v. ABTS (2,2′-Azinobis (3-ethylbenzothiazoline-6-sulfonic acid), potassium persulfate ($K_2S_2O_8$) and chromatographic solvents (methanol and acetonitrile) for analytical HPLC were purchased from Sigma (St Louis, MO, USA). DPPH (1,1-Diphenyl-2-picryl-hydrazyl) free radical were purchased from TCI (St Louis, MO, USA). L-ascorbic acid (≥ 98%) was purchased from Chengdu Kelong Chemical Reagent