



E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2015; 4(1): 29-31
Received: 20-02-2015
Accepted: 23-03-2015

Vladimir A. Kurkin
The Department of
Pharmacognosy with Botany
and Basis of Phytotherapy,
Samara State Medical
University, Ul. Chapaevskaya
89, Samara, 443099, Russia.

Saffloroside, a new flavonoid from flowers of *Carthamus tinctorius* L.

Vladimir A. Kurkin

Abstract

From the flowers of *Carthamus tinctorius* L., cultivated in Samara Region (Russian Federation) there was isolated a new flavonoid, named us saffloroside, which has the structure of 7-O- β -D-glucopyranoside of 3, 7, 3¹, 4¹-tetrahydroxy-5-methoxyflavone (azaleatin). The chemical structure of saffloroside there was elucidated by means of ¹H-NMR-, UV-spectroscopy and mass spectrometry. In the course of phytochemical investigations on the flowers of *Carthamus tinctorius* there were isolated also cynaroside and luteolin previously described for this plant.

Keywords: *Carthamus tinctorius* L., flowers, flavonoids, saffloroside, cynaroside, azaleatin, luteolin.

1. Introduction

Safflower (*Carthamus tinctorius* L., *Asteraceae*) is a famous oil-bearing culture, the seeds of which contains up to 40% of fatty oil, used as a component for the production of phytopharmaceuticals and cosmetic products [1]. Earlier we have studied the fatty-acid composition of fatty oil from the seeds of safflower [2]. However, the flowers of this plant are interesting as a source of phytopharmaceuticals, which have antioxidant, hepatoprotective, anti-inflammatory, neurotropic and other properties [3, 4]. In our opinion, from the point of view of antioxidant, hepatoprotective activities of preparations from flowers from safflower the flavonoids are of interest. According to reference data [3, 4, 5, 6]. The flowers of this plant contain acacetin, luteolin, quercetin, 7-O- β -D-glucuronoside of acacetin, 6-C- β -D-glucuronosyl-8-C- β -D-glucuronoside of acacetin, 7-O- β -D-glucopyranoside of luteolin (cynaroside), 7-O- β -D-glucuronoside of luteolin, luteolin-7-O-(6¹¹-O-acetyl)- β -D-glucuronoside, kaempferol-3-O-rutinoside (nicotiflorin), glycosides of 6-hydroxykaempferol, quercetin-3-O- β -D-glucuronoside, quercetin-7-O-7-O-(6¹¹-O-acetyl)- β -D-glucuronoside, quercetin-3-O-rutinoside (rutoside).

The aim of present work is the study flavonoid composition of safflower flowers, cultivated in the Samara region (Russian Federation).

2. Materials and methods

2.1. Plant materials: Flowers of *Carthamus tinctorius* L. were collected in Samara, Russian Federation, in August 2013, and identified by Dr. A.V. Milekhin, Samara Research Institute of Agriculture named of N.M. Tulaikov of Russian Agricultural Academy (Bezenchuk, Samara region). A voucher specimen (No. 573) is deposited at the Herbarium of the Department of Pharmacognosy, Samara State Medical University, Samara, Russian Federation.

2.2. Extraction and isolation: 100 g of air-dried flowers of safflower, collected in August 2013, was successively extracted with 70% EtOH, carrying out the first two extraction at room temperature for 24 hours, and then the extraction was carried out when heated to boiling water bath during 30 min. Combined water-alcoholic extract was evaporated under vacuum to a volume of 50 mL, mixed with 30 g of silica gel L 40/100 and dried. Dried powder (dry extract + silica gel) applied on a layer of silica gel (height of the layer of sorbent - 5 cm, diameter - 8 cm), formed as a suspension in chloroform. The chromatographic column was eluted with CHCl₃, and a mixture of CHCl₃-EtOH in different ratios (99:1; 98:2; 97:3; 95:5; 93:7; 90:10; 85:15; 80:20; 70:30; 60:40; 50:50). Fractions of 100 mL were collected and monitored by TLC analysis (Sorbfil PTLC-AF-A-UV, solvent systems CHCl₃-EtOH, 9:1; CHCl₃-EtOH-H₂O, 26:16:3; and *n*-butanol-glacial acetic acid-water, 4:1:2). Fractions containing a dominant compound **1** were combined, had fallen out of them precipitate was separated and crystallized

Correspondence:
Vladimir A. Kurkin
The Department of
Pharmacognosy with Botany
and Basis of Phytotherapy,
Samara State Medical
University, Ul. Chapaevskaya
89, Samara, 443099, Russia.

from water EtOH. At results from herbal material there were obtained 100 mg of flavonoid **1**. Fractions containing compound **2** were collected and after the evaporation they were drying on polyamide «Wolem». Dry powder (dry fractions + polyamide) rechromatographed on the chromatographic column (height of the layer of sorbent - 5.0 cm, diameter - 4 cm) using as eluent the solvent systems EtOH-H₂O (0:100, 20:80, 40:60, 70:30, 100:0). The fractions containing compound **2** (EtOH-H₂O (40: 60) were combined and then evaporated. At results there were obtained 30 mg of flavonoid **2**. Flavonoids **3** and **4** there isolated from *Carthamus tinctorius* flowers in the minor amounts.

2.3. Methods of structural elucidations. ¹H-NMR spectra of isolated compounds were obtained on the spectrometer of «Bruker AM 300» (300 MHz), mass spectra were taken on mass spectrometer «Kratos MS-30», UV-spectra were

registered on the spectrophotometer «Specord 40» (Analytik Jena). The acid hydrolysis of isolated glycosides **1** and **2** there were carried out by means of 2% HCl at 100 °C on the boiling bath during 30 min.

3. Results and discussion

3.1. Structural elucidations. At results of the phytochemical investigation, there were isolated from *Carthamus tinctorius* L. flowers two flavonoids (**1-4**) (Figure 1). At acid hydrolysis of isolated glycoside **1** there were obtained glucose and aglycone, identified on the basis of data of ¹H-NMR-, UV- and mass spectra as 3,7,3',4'-tetrahydroxy-5-methoxyflavone (azaleatin) (**3**) [7]. At acid hydrolysis of isolated glycoside **2** there were obtained glucose and aglycone, identified on the basis of data of ¹H-NMR-, UV- and mass spectra as 5,7,3',4'-tetrahydroxyflavone (luteolin) (**4**) [4, 8, 9, 10].

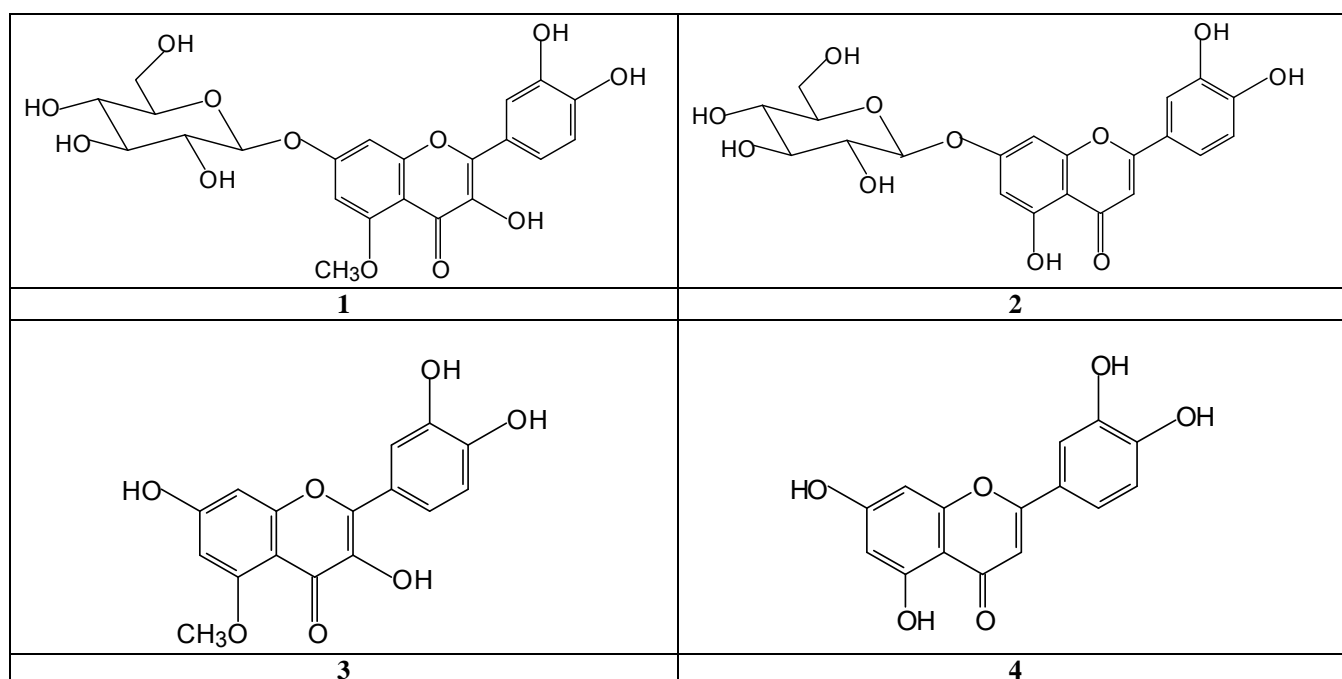


Fig 1: Chemical structures of flavonoids (1-4) isolated from *Carthamus tinctorius*.

In ¹H-NMR spectrum of compound **1** detected signals of aromatic protons H-2' at δ 7.78 (1H, d, J=2.5 Hz), H-6' at δ 7.75 (1H, dd, J=2.5 and 9 Hz), H-5' at δ 6.95 (1H, d, J=9 Hz), H-8 at δ 6.86 (1H, d, J=2.5 Hz) and H-6 at δ 6.43 (1H, d, J=2.5 Hz). Detection of singlet signal (3H) at δ 3.83 indicates the presence in the molecule of flavonoid **1** of the methoxy group, which is related to the ring A of flavonoid on the basis of data of mass spectrum: peak of ion with m/z 168 corresponding to the fragment (A+H)⁺. The choice of a place of methylation in favor of 5-OH group is made on the basis of data of ¹H-NMR spectrum, which not contains the singlet signal of 5-OH group in contrast cynaroside (**2**). This conclusion is confirmed by the data of the UV spectra of flavonoid **1** and its aglycone (**3**): the absence of the shift shortwave band of UV spectra of in the presence of NaOAc [10] and the appearance of this effect in the case of azaleatin (**3**). β -Configuration glycoside bond of flavonoid **1** is confirmed by the presence in ¹H-NMR spectrum of doublet signal (J=7 Hz) at δ 5.05 belonging to anomeric proton of glucose.

Flavonoid **2** on the basis of data of UV, ¹H-NMR and mass spectra there was identified us as 7-O- β -D-glucopyranoside of 5, 7, 3', 4'-tetrahydroxyflavone (cynaroside) [4].

In the course present investigation, there was isolated from *Carthamus tinctorius* L. flowers 7-O- β -D-glucopyranoside of 3,7,3',4'-tetrahydroxy-5-methoxyflavone (saffloroside) (**1**), which is a new natural compound. It is important to note, that saffloroside (**1**) is predominant flavonoid of *Carthamus tinctorius* L. flowers. The second flavonoid in importance is the cynaroside (7-O- β -D-glucopyranoside of 5, 7, 3', 4'-tetrahydroxyflavone) (**2**), previously described for this plant [3]. In addition, we have not found the acacetin, kaempferol, 6-hydroxykaempferol, quercetin and their glycosides previously described for this plant [3, 4, 5, 6].

In our opinion, saffloroside (**1**) may be of interest in terms of standardization of herbal materials, including identification of *Carthamus tinctorius* L. flowers.

3.2. Physical-chemical and spectral characteristics of isolated compounds

Saffloroside: Compound **1** was isolated as yellow crystals (water EtOH); mp 230-233 °C; C₂₂H₂₂O₁₂, R_f 0.50, Sorbfil PTLCAF-A-UV, CHCl₃/EtOH/H₂O (26:16:3); UV spectrum (EtOH, λ_{max} , nm): 260, 274sh, 371 nm; + NaOAc 260, 274sh, 371 nm; + NaOAc + H₃BO₃ 260, 378 nm; +AlCl₃ 274, 432

nm; MS-EI (70 eV, 200 °C, m/z, %): 316 (M⁺ of aglycone, 100 %), 168 (2), 153 (12), 137 (8); ¹H NMR spectrum (300 MHz, DMSO-d₆, δ, ppm, J/Hz): 7.78 (1H, d, J = 2.5, H-2¹), δ 7.75 (1H, dd, J = 2.5 and 9, H-6¹), 6.95 (1H, d, J = 9, H-5¹), 6.86 (1H, d, J = 2.5, H-8), 6.43 (1H, d, J = 2.5, H-6), 5.05 (1H, d, J = 7, H-1¹¹), 3.83 (3H, s, CH₃O), 3.8 -3.1 (6H, m, Glc).

Cynaroside: Compound **2** was isolated as light-yellow crystals (water EtOH); mp 232-234°C; C₂₁H₂₀O₁₁, R_f 0.45, Sorbfil PTLC-AF-A-UV, CHCl₃/EtOH/H₂O (26:16:3); UV spectrum (EtOH, λ_{max}, nm): 257, 266sh, 352 nm; +NaOAc 258, 268sh, 380 nm; +AlCl₃ 276, 330, 350, 394 nm; MS-EI (70 eV, 200 °C, m/z, %): 286 (M⁺ of aglycone, 100%), 153 (52), 137 (46); ¹H NMR spectrum (300 MHz, DMSO-d₆, δ, ppm, J/Hz): 12.98 (1H, s, 5-OH group), 7.45 (2H, dd, J = 2 and 9, H-6¹), 7.41 (1H, d, J = 2, H-2¹), 6.91 (1H, d, J = 9, H-5¹), 6.78 (1H, d, J = 2, H-8), 6.73 (1H, d, J = 2, H-6), 6.42 (1H, s, H-3), 5.03 (1H, d, J = 7.2, H-1¹¹), 3.9 - 3.1 (6H, m, Glc).

3, 7, 3¹, 4¹-tetrahydroxy-5-methoxyflavone (azaleatin): Compound **3** was isolated as yellow amorphous powder; C₁₆H₁₂O₇, R_f 0.90, Sorbfil PTLC-AF-A-UV, CHCl₃/EtOH/H₂O (26:16:3); UV spectrum (EtOH, λ_{max}, nm): 260, 274sh, 371 nm; + NaOAc 265, 277sh, 372 nm; + NaOAc + H₃BO₃ 260, 378 nm; +AlCl₃ 274, 432 nm. MS-EI (70 eV, 200 °C, m/z, %): 316 (M⁺, 100 %), 168 (8), 167 (25), 137 (34). ¹H-NMR spectrum (300 MHz, DMSO-d₆, δ, ppm, J/Hz): 7.78 (1H, d, J = 2.5, H-2¹), 7.75 (1H, dd, J = 2.5 and 9, H-6¹), 6.95 (1H, d, J = 9, H-5¹), 6.86 (1H, d, J = 2.5, H-8), 6.43 (1H, d, J = 2.5, H-6), 3.82 (3H, s, CH₃O).

5,7,3¹,4¹-tetrahydroxyflavone (luteolin): Compound **4** was isolated as yellow crystals; mp 282-285 °C (water EtOH); C₁₅H₁₀O₆, UV spectrum (EtOH, λ_{max}, nm): 256, 266sh, 358 nm; +NaOAc 259, 268sh, 390 nm; +AlCl₃, 278, 330, 355, 404 nm. MS-EI (70 eV, 200 °C, m/z, %): 286 (M⁺, 100%), 153 (17), 137 (30). ¹H-NMR spectrum (300 MHz, DMSO-d₆, δ, ppm, J/Hz): 12.60 (1H, s, 5-OH group), 7.40 (1H, dd, J = 2 and 9, H-6¹), 7.37 (1H, d, J = 2, H-2¹), 6.88 (1H, d, J = 9, H-5¹), 6.65 (s, H-3), 6.42 (1H, d, J = 2, H-8), 6.18 (1H, d, J = 2, H-6).

4. Conclusion

From the flowers of *Carthamus tinctorius* L., cultivated in Samara Region (Russian Federation) there was isolated saffloroside, a new flavonoid, which has the structure of 7-O-β-D-glucopyranoside of 3, 7, 3¹, 4¹-tetrahydroxy-5-methoxyflavone (azaleatin). In the course of phytochemical investigation on the flowers of *Carthamus tinctorius* there were isolated also cynaroside and luteolin previously described for this plant.

5. Acknowledgement

Thanks are due to Dr. Sci. Elena D. Daeva, Zelinsky Institute of Organic Chemistry of the Russian Academy of Sciences, Moscow, Russian Federation for measuring of NMR spectra and to Dr. Sci. Valentin I. Kadentsev, Zelinsky Institute of Organic Chemistry of the Russian Academy of Sciences, Moscow, Russian Federation for measuring of mass spectra.

6. Reference

1. Shikov AV, Makarov VG, Ryzhenkov VE. Plant oils and oil extracts: technology, standardization, properties. Moscow. Russian Doctor, 2004.

- Kurkin VA, Kharisova AV, Platonov IA, Pavlova LV, Milekhin AV. The study of composition of fatty acids of seed oils of *Carthamus tinctorius*. Pharmacy 2013; 61(5):30-32.
- Yoon HR, Han HG, Pa YS. Flavonoid Glycosides with Antioxidant Activity from the Petals of *Carthamus tinctorius*. J. Appl. Biol. Chem. 2007; 50(3):175-178.
- Yue S, Tang Yu, Li S, Duan JA. Chemical and Biological Properties of Quinochalcone C-Glycosides from the Florets of *Carthamus tinctorius*. Molecules 2013; 18:15220-15254.
- Hattori M, Huang XL, Che QM, Kawata Y, Tezuka Y, Kikuchi T *et al.* 6-hydroxykaempferol and its glycosides from *Carthamus tinctorius* petals Phytochemistry 1992; 31(11):4001-4004.
- Meselhy MR, Kadota S, Momose Y, Hatakeyama N, Kusai A, Hattori M *et al.* Two new quinochalcone yellow pigments from *Carthamus tinctorius* and Ca²⁺ antagonistic activity of tinctormine. Chem. Pharm. Bull. 1993; 41:1796-1802.
- Dinda B, Das SK, Hajra AK, Bhattacharya A, De K, Chel G *et al.* Indian Journal of Chemistry 1999; 38B:577-582.
- Harborne JB, Mabry TJ, Mabry H. (Eds.) The Flavonoids. London. Chapman & Hall, 1975.
- Harborne JB, Mabry TJ. (Eds.) The Flavonoids: Advances in Research. London; New York. Chapman & Hall, 1982.
- Mabry TJ, Markham KR, Thomas MB. The Systematic Identification of Flavonoids. Berlin-Heidelberg-New York. Springer Verlag, 1970.