ORIGINAL ARTICLE

A C G publications

Rec. Nat. Prod. X:X (2020) XX-XX

records of natural products

A New Cynaropicrin Derivative from *Cynara Scolymus* L.

Ghada M. Abbas[1,2], Amal Sallam[1], Fatma M. Abdel Bar[1,3], Mohamed Farid I. Lahloub[1] and Ahmed A. Gohar[1*]

[1]*Department of Pharmacognosy, Faculty of Pharmacy, Mansoura University, Mansoura 35516, Egypt*
[2]*Department of Pharmacognosy, Faculty of Pharmacy, Horus University in Egypt (HUE), New Damietta, 34517, Egypt*
[3]*Department of Pharmacognosy, Faculty of Pharmacy, Prince Sattam Bin Abdulaziz University, Al-Kharj 11942, Saudi Arabia*

(Received July 24, 2020; Revised August 27, 2020; Accepted August 28, 2020)

Abstract: A new sesquiterpene lactone **1a** along with other four known compounds **1b**, **2**, **3** and **4** were isolated from fresh leaves of *Cynara scolymus* L. using ordinary chromatographic techniques. The structures of the isolated compounds were determined via spectroscopic analysis.

Keywords: *Cynara scolymus*; sesquiterpene lactones; dihydrocynaropicrin; tetrahydrocynaropicrin; pinoresinol; luteolin**.**

1. Introduction

Globe artichoke (*Cynara cardunculus* var. *scolymus* Fiori), formerly *Cynara scolymus* L., Asteraceae family, is an ancient herbaceous perennial plant, originating from the Mediterranean area. It's cultivated, worldwide, for its leaves, the large immature capitula with edible fleshy bracts and receptacle [1]. In addition of being consumed as a food, *C. scolymus* is recognized as herbal medicine [2]. Leaves which are rich in polyphenols are mostly utilized in nutraceuticals for the production of commercial extracts [3]. Flower heads and roots are used as prebiotic ingredient in functional foods due to their inulin content [3]. Several health promoting effects of artichoke extracts have been demonstrated including hepatoprotective [4], choleretic [5], anticholestatic [6], antioxidative [7], hypolipidemic [8], antispasmodic [9], antimicrobial [10] and antihypercholesterolemic effects [11]. Additionally, artichoke extracts have shown antitumor [12], analgesic and anti-inflammatory [13] activities. Phytochemical studies of *C. scolymus* revealed the presence of several classes of compounds such as caffeoylquinic acids derivatives, sesquiterpene lactones, flavonoids, saponins, phenolic acids [14] and the sesquiterpene glycosides; cynarascolosides A, B, and C [8]. Whereas, anthocyanins were present only in the capitula of this plant [15]. Chlorogenic acid and 1,5-dicaffeoylquinic acids are the predominant compounds among hydroxycinnamates of *C. scolymus* [2]. A published review on the sesquiterpene lactone cynaropicrin, indicated that it contributes to approximately 80% of the characteristic bitterness of artichoke, reported to have vital biological activities as anti-HCV, antiparasitic, antihyperlipidemic, antifeedant, antiphotoaging, antioxidant, antibacterial, anti-inflammatory, anti-gastritis and antitumor [16].

* Corresponding author: E-Mail: ahmedgohar99@yahoo.com; Phone: +2 01008701009 *Fax:* 050-2200242

The article was published by ACG Publications
http://www.acgpubs.org/journal/records-of-natural-products Month-Month 2020 EISSN:1307-6167
DOI: http://doi.org/10.25135/rnp.198.20.07.1747

Owing to the valuable biological activities of *C. scolymus*, this study aims to revisit this plant to explore the presence of additional sesquiterpene lactones and other constituents that may contribute to such biological activities.

2. Materials and Methods

2.1. General

Rotary flash evaporator (Büchi, Switzerland), UV lamp 254 and 366 nm, Desaga (Germany), UV-visible spectrophotometer, Shimadzu 1601 PC, model TCC240 (Shimadzu, Kyoto, Japan), Mass spectrometry (HRAM/MS) spectra were obtained using a Thermo Scientific UPLC RS Ultimate 3000-Q Exactive hybrid quadrupole-Orbitrap mass spectrometer combines high-performance quadrupole precursor selection with high-resolution, accurate-mass (HR/AM) Orbitrap™ detection, Infra-red spectrophotometer, Thermo Scientific Nicolet™ iS™10 FT-IR Spectrometer (Thermo Fisher scientific Co., Madison, WI, USA), Thin-layer chromatography (TLC) was performed using pre-coated silica gel 60 GF_{254} (20 x 20 cm, 0.2 mm thick) on aluminum sheets (Merck, Darmstadt, Germany) and pre-coated reversed phase C_{18} silica gel glass plates, Partisil® $KC_{18}F$ Silica gel 60A with fluorescent indicator (5 x 20 cm, 200 µm layer thickness). Vanillin-sulfuric acid (5%), 5% aluminum chloride, ferric chloride spray reagents. Nuclear magnetic resonance spectra (^{1}H NMR, ^{13}C NMR, APT and other 2D spectra) were recorded with Bruker Avance III spectrophotometer at 400 MHz for proton and at 100 MHz for carbon. For column chromatography; glass columns of different dimensions were used, normal phase chromatography was carried out using silica gel G 60-230 (Merck, Darmstadt, Germany) packed by the wet method in the stated solvent, gel permeation chromatography was carried out using Sephadex LH-20 (SIGMA-ALDRICH, Missouri, USA) in a medium pressure column and reversed phase chromatography was performed using phase-bonded octadecylsilyl-silica gel (RP-C_{18}, BAKERBOND® Octadecyl, C_{18}) 40 µm, Prep LC Packing (Phillipsburg, NJ, USA).

2.2. Plant Material

The plant material was collected in February 2017, from Agricultural Research Center farm, Dakahlia, Egypt. The plant identity was confirmed by Prof. Dr. Ibrahim Mashaly, Department of Botany, Faculty of Science, Mansoura University, Egypt. A voucher specimen 02-17-CS-Mansoura was deposited at the Herbarium of Faculty of Pharmacy, Mansoura University. The leaves were collected, milled and kept for phytochemical investigations.

2.3. Extraction and Isolation

In the present study, the CH_2Cl_2 fraction (110 g) of MeOH extract was coarsely fractionated over a silica gel column (20 x 4.5 cm i.d., 215 g) using gradient elution with petroleum ether-EtOAc, mixtures up to 100 v/v EtOAc. The effluents, 250 mL each, were collected, concentrated and similar fractions were combined into 17 groups. Four selected groups were used for the isolation of compounds **1- 4** as outlined in Figure 1.

Fractions 41-54 (Gr.1), 486 mg, eluted with petroleum ether-EtOAc (70: 30 v/v) displayed 3 major spots, R_f 0.71, 0.57 and 0.51, using MeOH-water (6: 4 v/v) as a solvent system and RP-C_{18} silica gel glass plates. The fraction was rechromatographed over a RP-C_{18} column (15 x 4.5 cm i.d., 200 g) and eluted with gradient elution with MeOH-H_2O mixtures starting with MeOH-H_2O (40: 60 v/v) till 100 % MeOH. The effluents, 10 mL fraction each, were collected and screened by TLC. Similar fractions were collected. Sub-fractions 2-3 (Gr.1A), 320 mg, eluted with MeOH- H_2O (40: 60 v/v), displayed three spots, R_f 0.19, 0.32 and 0.43, using CH_2Cl_2- MeOH (9.5: 0.5 v/v) as a solvent system and normal silica gel plate, two of them are major spots. The collected fraction was rechromatographed over normal silica column (35 x 2 cm i.d., 200 g) and eluted with gradient elution with CH_2Cl_2- MeOH

mixtures, (100: 0 v/v) till (0: 100 v/v). The effluents (20 mL) were collected and screened by TLC. Similar fractions were collected; sub-fractions 68-83, 64.4 mg, eluted with CH_2Cl_2- MeOH (50: 50 v/v) were collected and further purified over normal silica gel column (18 x 0.7 cm i.d., 2.5 g) and gradient-eluted with CH_2Cl_2- EtOAc mixtures, (100: 0 v/v) till (0: 100 v/v). The effluents (100 mL) were collected and screened by TLC. Similar fractions were collected; sub-fractions 27-38, eluted with CH_2Cl_2- EtOAc (7: 3 v/v), were collected and further purified over Sephadex LH20 column using isocratic elution with 100% CH_2Cl_2 to afford compound **2** (Fr.1- 6), 40 mg.

Sub-fractions 9-11 (Gr.1B), 55 mg, eluted with MeOH- H_2O mixtures (40: 60 v/v) displayed one major spot, R_f 0.29 using MeOH- H_2O mixtures (5: 5 v/v) as a solvent system and C_{18} silica gel glass plates. The fraction was re-purified on RP-C_{18} column using gradient elution starting with MeOH-H_2O (30: 70 v/v) till (33: 67 v/v) and fractions of 10 mL were collected to afford compound **1** (sub-fractions 21-53), 45 mg. Sub-fractions 12-20 (Gr.1C), 32 mg, eluted with MeOH- H_2O (40: 60 v/v), were re-chromatographed over normal silica column (16 x 0.7 cm i.d., 2 g) and gradient-eluted with CH_2Cl_2- MeOH mixtures, (100: 0 v/v) to (80: 20 v/v). The effluents, 10 mL fraction each, were collected and screened by TLC. Similar fractions were collected; sub-fraction 32, 8 mg, eluted with CH_2Cl_2-MeOH (9.5: 0.5 v/v), was further purified over normal silica column (5 x 0.5 cm i.d., 0.5 g) and eluted with gradient elution with CH_2Cl_2-EtOAc mixtures started with 100% CH_2Cl_2 till (95: 5 v/v). The effluents (1 mL) were collected and screened by TLC. Similar fractions were collected; sub-fractions 40-73, eluted with CH_2Cl_2- EtOAc (96: 4 v/v) were collected to afford compound **3**, 4.5 mg.

Sub-fractions 34-40 (Gr.1D), 60 mg, eluted with MeOH- H_2O (50: 50 v/v), displayed one major spot, R_f 0.63 using CH_2Cl_2- MeOH (9.5: 0.5 v/v) as a solvent system and normal silica gel plate and acquired yellow color after heating with vanillin/sulfuric acid. The formed precipitate washed three times with CH_2Cl_2- MeOH mixture (5:5 v/v) to afford compound **4**, 40 mg.

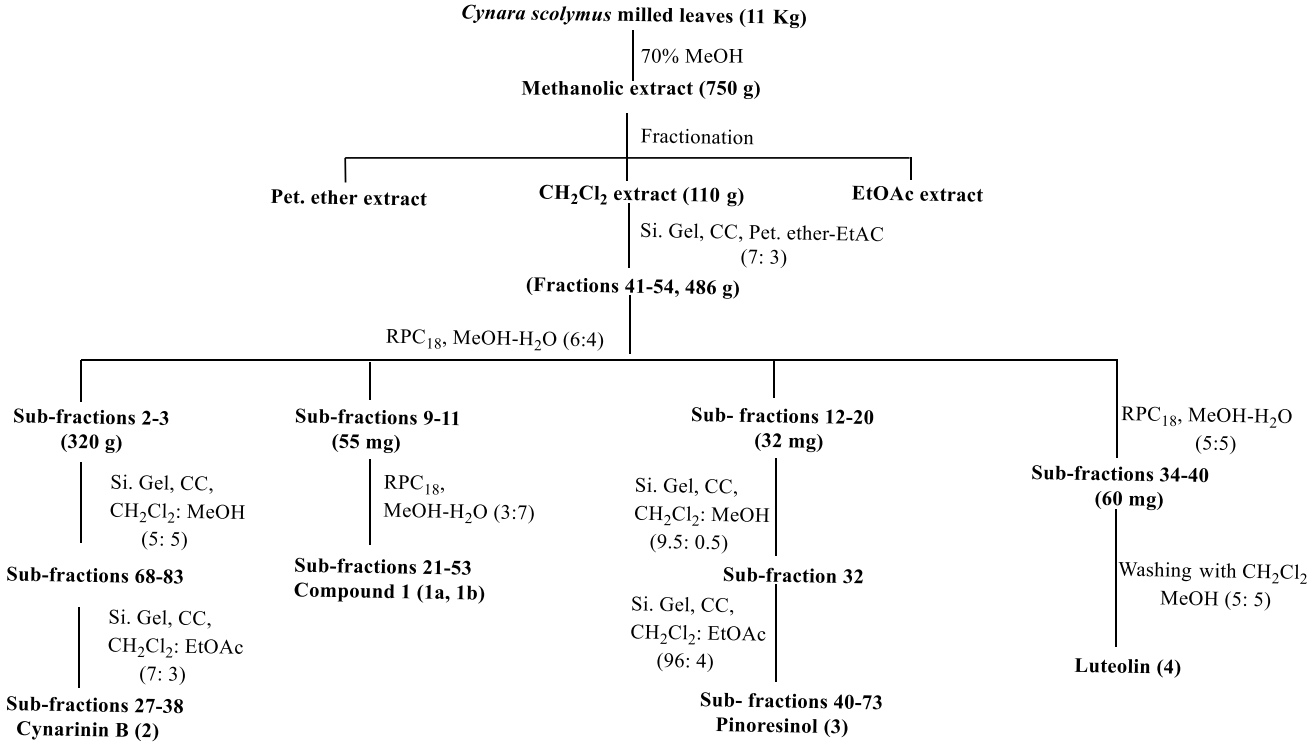

Figure 1. Isolation procedures of compounds **1- 4**

We were able to identify a new cynaropicrin derivative (**1a**) from the CH_2Cl_2 fraction of the methanol extract of fresh *C. scolymus* leaves. This new derivative was isolated as a mixture with the known sesquiterpene lactone, 11,13-dihydrocynaropicrin (**1b**) [17]. Different techniques were used in isolation including repeated column chromatography using silica gel, RP-C_{18} or Sephadex LH-20. Extensive 1D and 2D NMR spectroscopic analyses were applied for the structural elucidation.

Moreover, the isolation procedures afforded three other known compounds; the sesquiterpene lactone cynarinin B (**2**), the lignan pinoresinol (**3**) and the flavonoid luteolin (**4**).

3. Results and Discussion

3.1. Structure Elucidation

Compound **1** (20 mg) was obtained as a transparent to slightly yellowish viscous oil. Careful examination of the ^{1}H- and APT spectra and correlating the data from them with that obtained from both HSQC and HMBC suggested the presence of three methyls, eleven methylenes, thirteen methines, and nine quaternary carbons, two of them are γ-lactone carbonyls. This data suggests that **1** is a mixture of two sesquiterpene lactones **1a** and **1b**. This was further supported by data obtained from HRMS that showed clear two molecular ion peaks at *m/z* 395.1710 and 393.1550 $[M+ HCOO^{\bullet}]^{-}$ (calcd. 395.1706 and 393.1549), respectively. So, we suggested that one of the derivatives is the dehydro form of the other one and this is supported by the dehydration process occurring during the biosynthetic pathway of this series of compounds due to activity of NADP enzyme [18]. Data from 1D and 2D spectra revealed that one of the two compounds (**1a**, Figure 2) possesses 19 carbons established for two methyls, five methylenes, seven methines, and four quaternary carbons. The resonance at δ_c 176.6 was assigned to a γ-lactone carbonyl carbon (C-12). The two methyl signals at δ_c 15.4 and 18.3 corroborated to the proton signals at δ_H 1.25 (3H, s) and 1.19 (3H, *br.s*) were assigned by HSQC experiment (Figure S10) and further confirmed by HMBC experiment (Figure 2 and S12) to CH_3-13 and CH_3-15, respectively. The ^{1}H NMR spectrum displayed two broad singlets at δ_H 5.10 (1H) and 5.11 (1H) attributable to a pair of vinylic hydrogens of an exomethylene group (H_2-14) which were assigned to the carbon resonance at δ_c 116.3 (C-14), based on HSQC correlation with C-14 and HMBC correlations between these protons (i.e. H_2-14) and carbon signals at δ_c 42.3 (C-1) and 42.4 (C-9). Three oxygenated carbons were displayed by APT spectrum at δ_c 78.0, 81.0 and 76.9, were assigned to C-3, C-6 and C-8, respectively. These spectroscopic data were consistent with those of a guaianolide-type sesquiterpene lactone. The downfield shift of H-8 at δ_H 4.94 (1H, *br.s*) and the deshielded C-8 at δ_C 76.9 evidenced the acylation of the hydroxyl group at this position [19]. The presence of four carbon resonances at δ_C 165.2 (C=O), 139.9 (C), 126.3 (CH_2) and 62.0 (CH_2OH) were assigned to C-16, C-17, C-19 and C-18, respectively of the acyl moiety at C-8. HSQC spectrum correlated the proton singlet at δ_H 4.35 (2H) to the hydroxymethylene C-18 and the broad singlet at δ_H 5.94 (2H) to the exomethylene C-19. The proton signal H_2-18 showed HMBC correlations with C-16 and C-17, whereas the proton signal H_2-19 showed HMBC correlations with C-16, C-17 and C-18 (Figure 2) confirming their assignments. Therefore, the ester's side chain at C-8 was defined as 4-hydroxymethacrylate.

The stereochemical configuration was related to cynaopicrin [20, 21], meanwhile the NOESY correlations allowed the determination of H-13 and 1-15 orientation. NOESY correlations (Figure 3) between H-11 at δ_H 2.50 (1H, m) with H-8 δ_H 4.94 (1H, *br.s*) and H-6 δ_H 4.04 (1H, *t*, *J*= 8.0) allowed the determination of CH_3-13 as *α*-oriented. NOESY correlation between H-3 at δ_H 3.71 (1H, *br.s*) with H-1 at δ_H 2.62 (1H, *m*), H-5 at δ_H 1.95 (1H, *m*) and CH_3-15 at δ_H 1.19 (3H, *br.s*), allowed the determination of CH_3-15 as *α*-oriented. The absence of NOESY correlations (Figure 3) between H-6 at δ_H 4.04 (1H, *t*, *J*= 8.0 Hz) and both of H-5 at δ_H 1.95 (1H, *m*) and H-7 at δ_H 2.20 (1H, m) indicated that the cycloheptene ring and the γ-lactone ring were *trans*-fused. The spectral data of **1a** were highly similar to the previously reported data for cynaropicrin [22, 23] except for the methyl substitutions at C-4 and C-11 instead of exomethylene groups in cynaropicrin.

Based on the collective spectral data of the residual peaks after subtracting the identified peaks, the second compound **1b** was almost similar to those published for a semi-synthesized cynaropicrin derivative [17]. Careful examination of spectral data of **1b** confirmed the presence of guaianolide-type sesquiterpene lactone nucleus with a 4-hydroxymethacrylate side chain at δ_C 76.3 (C-8), two exomethylene groups at δ_C 112.4 (C-15) and 117.4 (C-14), a hydroxyl group at δ_C 73.5 (C-3) and a carbon resonance at δ_C 15.4 (C-13) assigned to the methyl group at C-11. The vinylic hydrogens of the exomethylene group CH_2-14 was assigned to the overlapped signal at δ_H 5.03, and the broad singlet at δ_H 5.14 as revealed from HSQC spectrum and from ^{3}J-HMBC correlation with C-1 (δ_C 44.2). Additionally, the methylene carbon at δ_c 112.4, correlated in HSQC spectrum with a pair of broad

singlets at δ_H 5.33 and 5.40 (H_2-15), showed HMBC correlation with carbon signal at δc 73.5 (C-3) confirming its assignment.

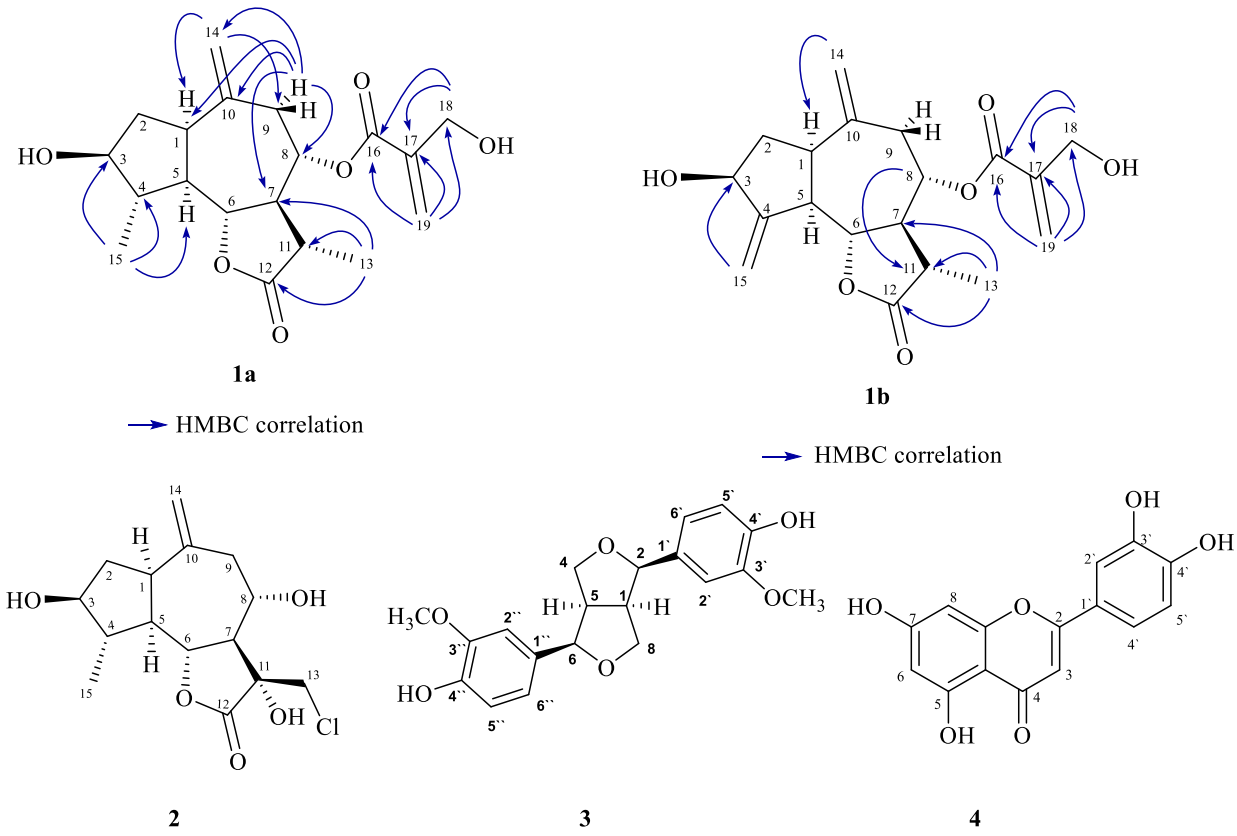

Figure 2. Structure of the isolated compounds from *Cynara scolymus* L.

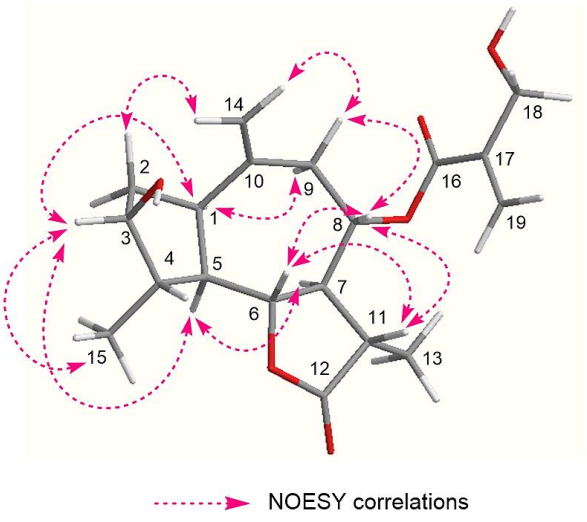

Figure 3. Significant NOESY correlations of **1a**

From the previous data and by comparison with the reported ones, compound **1** was confirmed to be a mixture of two sesquiterpene lactones; the first (**1a**) is 4,11,13,15-tetrahydrocynaropicrin, which is a new compound and the second one (**1b**), 11,13-dihydrocynaropicrin, is a known one that was

previously semi-synthesized from cynaropicrin [17] and is reported here for the first time as a natural compound. The structures of the remaining compounds **2**, **3** and **4** (Figure 2) were elucidated by comparing their ^{1}H and ^{13}C NMR spectral data with those reported in the literature as cynarinin B (**2**) [24], pinoresinol (**3**) [25] and luteolin (**4**) [26].

Table 1. APT (at 100 MHz) and ^{1}H NMR (at 400 MHz) data for compounds **1a** and **1b** in $CDCl_3$

C/H No.	1a		1b	
	APT, δ_C in ppm	**δ_H in ppm, *J* in Hz**	**APT, δ_C in ppm**	**δ_H in ppm, *J* in Hz**
1	42.3 (CH)	2.81 (1H, *m*)	44.2 (CH)	2.92 (1H, *m*)*
2	38.1 (CH_2)	1.74 (1H, *m*, 2*β*)*	38.7 (CH_2)	1.75 (1H, *m*, 2*β*)*
		2.12 (1H, *m*, 2*α*)*		2.31 (1H, *m*, 2*α*)
3	78.0 (CH)	3.71 (1H, *br.s*)	73.5 (CH)	4.54 (1H, *br.s*)
4	47.0 (CH)	1.89 (1H, *br.s*)	152.6 (CH)	1.89 (1H, *br.s*)
5	50.9 (CH)	1.95 (1H, *m*)	50.5 (CH)	2.82 (1H, *m*)
6	81.0 (CH)	4.04 (1H, *t*, *J*= 8.0)	78.9 (CH)	4.16 (1H, *t*, *J*= 8.0)
7	55.8 (CH)	2.20 (1H, *m*)	53.3 (CH)	2.29 (1H, *m*)
8	76.9 (CH)	4.94 (1H, *br.s*)	76.3 (CH)	5.03 (1H, *br.s*)
9	42.4 (CH_2)	2.18 (1H, *m*, 9*α*)*	40.3 (CH_2)	2.27 (1H, *m*, 9*α*)*
		2.82 (1H, *m*, 9*β*)*		2.75 (1H, *m*, 9*β*)*
10	142.0 (C)	---	142.0 (C)	---
11	41.4 (CH)	2.50 (1H, *m*)	41.3 (CH)	2.50 (1H, *m*)
12	176.6 (C)	---	178.0 (C)	---
13	15.4 (CH_3)	1.25 (3H, *s*)	15.4 (CH_3)	1.25 (3H, *s*)
14	116.3 (CH_2)	5.11 (1H, *br.s*, 14b)	117.4 (CH_2)	5.14 (1H, *br.s*, 14b)
		5.10 (1H, *br.s*, 14a)		5.03 (1H, *br.s*, 14a)
15	18.3 (CH_3)	1.19 (3H, *br.s*)	112.4 (CH_2)	5.33 (1H, *br.s*, 15b)
				5.40 (1H, *br.s*, 15a)
16	165.2 (C)	---	165.2 (C)	---
17	139.3 (C)	---	139.3 (C)	---
18	62.0 (CH_2)	4.35 (2H, *br.s*)	62.0 (CH_2)	4.35 (2H, *br.s*)
19	126.3 (CH_2)	5.94 (2H, *br.s*)	126.3 (CH_2)	6.27 (2H, *br.s*)

*Overlapping signals

Supporting Information

Supporting information accompanies this paper on http://www.acgpubs.org/journal/records-of-natural-products

ORCID
Ghada M. Abbas: 0000-0003-4545-3954
Amal Salam: 0000-0003-3577-2046
Fatma M. Abdel Bar: 0000-0002-4690-4975
Mohamed Farid I. Lahloub: 0000-0001-6769-3145
Ahmed A. Gohar: 0000-0001-5906-1543

References

[1] V. Lattanzio, P. A. Kroon, V. Linsalata and A. Cardinali (2009). Globe artichoke: A functional food and source of nutraceutical ingredients, *J. Funct. Foods*. **1**, 131-144.

[2] S. Lombardo, G. Pandino, G. Mauromicale, M. Knödler, R. Carle and A. Schieber (2010). Influence of genotype, harvest time and plant part on polyphenolic composition of globe artichoke [*Cynara cardunculus* L. var. scolymus (L.) Fiori], *Food Chem.* **119**, 1175-1181.

[3] S. Raccuia and M. Melilli (2004). *Cynara cardunculus* L., a potential source of inulin in the Mediterranean environment: screening of genetic variability, *Aust. J. Agric. Res.* **55**, 693-698.
[4] G. Mehmetçik, G. Özdemirler, N. Koçak-Toker, U. Çevikbaş and M. Uysal (2008). Effect of pretreatment with artichoke extract on carbon tetrachloride-induced liver injury and oxidative stress, *Exp. Toxicol. Pathol.* **60**, 475-480.
[5] P. Matuschowski, A. Nahrstedt and H. Winterhoff (2005). Pharmacological investigations on the effect of fresh juice from *Cynara scolymus* on choleretic effects, *Z. Phytotherapie* **26**, 14-19.
[6] R. Gebhardt (2001). Anticholestatic activity of flavonoids from artichoke (*Cynara scolymus* L.) and of their metabolites, *Med. Sci. Mon. Int. Med. J. Exp. Clin. Res.* **7**, 316-320.
[7] R. Gebhardt and M. Fausel (1997). Antioxidant and hepatoprotective effects of artichoke extracts and constituents in cultured rat hepatocytes, *Toxicol. In Vitro* **11**, 669-672.
[8] H. Shimoda, K. Ninomiya, N. Nishida, T. Yoshino, T. Morikawa, H. Matsuda and M. Yoshikawa (2003). Anti-hyperlipidemic sesquiterpenes and new sesquiterpene glycosides from the leaves of artichoke (*Cynara scolymus* L.): structure requirement and mode of action, *Bioorg. Med. Chem. Lett*. **13**, 223-228.
[9] F. Emendörfer, F. Emendörfer, F. Bellato, V. F. Noldin, V. Cechinel-Filho, R. A. Yunes, F. Delle Monache and A. M. Cardozo (2005). Antispasmodic activity of fractions and cynaropicrin from *Cynara scolymus* on guinea-pig ileum, *Biol. Pharm. Bull.* **28**, 902-904.
[10] S. L. Castillo, N. Heredia, J. F. Contreras and S. García (2011). Extracts of edible and medicinal plants in inhibition of growth, adherence, and cytotoxin production of Campylobacter jejuni and *Campylobacter coli*, *J. Food Sci.* **76**, M421-M426.
[11] R. Gebhardt (2002). Inhibition of cholesterol biosynthesis in HepG2 cells by artichoke extracts is reinforced by glucosidase pretreatment, *Phytother. Res.* **16**, 368-372.
[12] V. F. Noldin, V. Filho, F. Monache, J. C. Benassi, I. L. Christmann, R. C. Pedrosa and R. A. Yunes (2003). Composição química e atividades biológicas das folhas de *Cynara scolymus* L.(alcachofra) cultivada no Brasil, *Quim. Nova.* **26**, 331-334.
[13] P. Trouillas, C.-A. Calliste, D.-P. Allais, A. Simon, A. Marfak, C. Delage and J.L. Duroux (2003). Antioxidant, anti-inflammatory and antiproliferative properties of sixteen water plant extracts used in the Limousin countryside as herbal teas, *Food Chem.* **80**, 399-407.
[14] I. M. Abu-Reidah, D. Arráez-Román, A. Segura-Carretero and A. Fernández-Gutiérrez (2013). Extensive characterisation of bioactive phenolic constituents from globe artichoke (*Cynara scolymus* L.) by HPLC–DAD-ESI-QTOF-MS, *Food Chem.* **141**, 2269-2277.
[15] K. Schütz, M. Persike, R. Carle and A. Schieber (2006). Characterization and quantification of anthocyanins in selected artichoke (*Cynara scolymus* L.) cultivars by HPLC–DAD–ESI–MS n, *Anal. Bioanal. Chem*. **384**, 1511-1517.
[16] M. F. Elsebai, A. Mocan and A. G. Atanasov (2016). Cynaropicrin: A comprehensive research review and therapeutic potential as an anti-hepatitis C virus agent, *Front. Pharmacol*. **7**, 472-472.
[17] G. Cravotto, G. M. Nano, A. Binello, P. Spagliardi and G. Seu (2005). Chemical and biological modification of cynaropicrin and grosheimin: a structure–bitterness relationship study, *J. Sci. Food Agric.* **85**, 1757-1764.
[18] P. A. Ramos, A. M. Ferro, M. M. Oliveira, S. Gonçalves, C. S. Freire, A. J. Silvestre and M. F. Duarte (2019). Biosynthesis and bioactivity of *Cynara cardunculus* L. guaianolides and hydroxycinnamic acids: a genomic, biochemical and health-promoting perspective, *Phytochem. Rev*. **18**, 495-526.
[19] A. Marzouk (2015). Investigating Volutaria abyssinica as a potential source for cytotoxic sesquiterpenoids, *J. Am. Sci*. **11**, 73-81.
[20] F. C. Seaman (1982). Sesquiterpene lactones as taxonomic characters in the asteraceae, *The Bot. Rev*. **48**, 121-594.
[21] A. Corbella, P. Gariboldi, G. Jommi, Z. Samek, M. Holub, B. Drożdż, and E. Błoszyk (1972). Absolute stereochemistry of cynaropicrin and related guaianolides, *J. Chem. Soc., Chem. Commun.* **7**, 386-387.
[22] N. Yayli, C. Baltaci, Y. Gök, E. Aydin and O. Üçüncü (2006). Sesquiterpene lactones from *Centaurea helenioides* Boiss, *Turk.J.Chem.* **30**, 229-233.
[23] T. J. Ha, D. S. Jang, J. R. Lee, K. D. Lee, J. Lee, S. W. Hwang, H. J. Jung, S. H. Nam, K. H. Park and M. S. Yang (2003). Cytotoxic effects of sesquiterpene lactones from the flowers of *Hemisteptia lyrata* B, *Arch. Pharm. Res.* **26**, 925-928.
[24] X. Li, P. Qian, Z. Liu, Y. Zhao, G. Xu, D. Tao, Q. Zhao, and H. Sun (2005). Sesquiterpenoids from *Cynara scolymus*, *Heterocycles* **65** (**2**), 287-291

[25] T. T. Diep, P. Van Kiem, N. T. Dong, N. H. Tung, B. T. Bang, C. Van Minh and A. Braca (2007). Pinoresinol and 3, 4', 5, 7-tetrahydroxy-3'-methoxyflavanone from the fruits of Silybum marianum (L.) Gaertn, *Vietnam J. Chem.* **45**, 219-222.
[26] M. A. Alwahsh, M. Khairuddean, and W. K. Chong (2015). Chemical constituents and antioxidant activity of *Teucrium barbeyanum* Aschers, *Rec. Nat. Prod.* **9**, 159-63.