Phytochemical Investigations on Chemical Constituents of *Taraxacum bessarabicum* (Hornem.) Hand.-Mazz. subsp. *bessarabicum* (Hornem.) Hand.-Mazz.

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**Abstract**

Plants of the genus *Taraxacum* Wigg., have long been used as medicinal herbs. A phytochemical investigation of the aerial parts of *Taraxacum bessarabicum* (Hornem.) Hand.-Mazz. subsp. *bessarabicum* (Hornem.) Hand.-Mazz. (Asteraceae) yielded two coumarins [esculetin (1), cichoriin (2)], three flavonoids [luteolin (3), luteolin 7-O-β-D-glucoside (4), gossypetin (5)] and six phenolic acids and their derivatives [p-coumaric acid (6), caffeic acid (7), ferulic acid (8), chlorogenic acid methyl ester (9), 3,5-di-O-caffeoylquinic acid (10), 3,5-di-O-caffeoylquinic acid methyl ester (11)]. Their structures were established conclusively by UV, ESI-MS, 1-D and 2-D NMR spectra analyses and comparison with literature data. The presence of these compounds has been shown for the first time from this species. This is the first report of the isolation of compound 5 from the genus *Taraxacum*.

**Keywords**: *Taraxacum bessarabicum* subsp; *bessarabicum*; Asteraceae; Coumarins; Flavonoids; Phenolic acids.

**Introduction**

The genus *Taraxacum* Wigg., commonly known as dandelion, belongs to the family Asteraceae, subfamily Cichorioideae, tribe Lactuceae (1-2). The name is derived from the Greek words ‘taraxis’, for inflammation, and ‘akeomai’ for curative (1). This plant genus has about 2500 species worldwide, and is widely distributed in the warmer temperature zones of the northern hemisphere (2-3). The total number of *Taraxacum* in Turkey at present is 43 species (4).

There are about 35,000 to 70,000 plant species that have been used for medicinal purposes worldwide (5). The plants of the genus *Taraxacum* have long been used in many traditional and modern herbal medical systems (1, 6).

The first evidence for its therapeutic use was mentioned by Arabian physicians of the 10th and 11th centuries to treat liver and spleen ailments (1). *Taraxacum* species have been traditionally used to medicate hepatic disorders, diarrhea, viral infections, anorexia, gout, and some women’s diseases, such as breast and uterus cancers, and as lactating, choleric, diuretic, and anti-inflammatory remedies (1, 5-6). In Turkish popular medicine, the plants of the genus *Taraxacum* are used as antirheumatic, anti-inflammatory, anti-diabetic medicines, for the treatment of eye diseases, stomach disorders, and kidney stones (9). Apart from being used as a pharmaceutical, the inflorescences, leaves...
and roots of *Taraxacum* species are processed into different food products. Young leaves of cultivated or wild species are consumed fresh as salad, whereas the roots are roasted and utilized as a coffee substitute. Additionally, the extracts are used as flavor components in various food products, including alcoholic beverages and soft drinks, candy, baked goods, gelatins and puddings, and cheese (1).

Previous phytochemical investigations have shown that *Taraxacum* species contain sesquiterpene lactones, triterpenes, phytosterols, flavonoids, lignans, coumarins, phenolic acids, beta-carbolin alkaloids, indole alkaloids, and carotenoids (1, 10-18).

*Taraxacum bessarabicum* (Hornem.) Hand.-Mazz. subsp. *bessarabicum* (Hornem.) Hand.-Mazz. is a perennial herbaceous plant. It is distributed mainly inner Anatolia and grows on saline places, fields, and 900-3000 m altitude (2). The isolation of sesquiterpene lactones and two phenolics has previously been reported from the roots of *Taraxacum bessarabicum* (11). In the present study, the aerial parts of *T. bessarabicum* subsp. *bessarabicum* collected from Erzincan, East Anatolia were investigated to elucidate their secondary-metabolite profile.

**Experimental**

**General experimental procedures**

UV Spectra: Shimadzu UV-1700 (PharmaSpec) spectrophotometer, in MeOH; λ<sub>max</sub> in nm. ¹H- and ¹³C-NMR Spectra: Varian Unity Inova 500 MHz spectrometer, at 500/125 MHz, resp., in CD<sub>3</sub>OD, δ in ppm rel. to Me<sub>4</sub>Si, J in Hz. ESIMS: Finnigan LCQ Advantage Max mass spectrometer, in m/z. Column chromatography (CC): silica gel 60 (40 – 63 μm; Merck) or Sephadex LH-20 (Sigma-Aldrich). Analitical and preparative TLC were performed on silica gel 60 F<sub>254</sub> plates (0.25 and 0.50 mm, respectively; Merck). Spots were visualized by exposure to UV radiation, NH<sub>3</sub> vapour, and NA spray reagent (Naturstoff reagenz-A). All solvents and chemical reagents were purchased from Merck (Darmshtot, Germany).

**Esculetin (1):** White powder; UV λ<sub>max</sub> (MeOH, nm): 221, 252 sh, 292, 344; ¹H-NMR (CD<sub>3</sub>OD, 500 MHz): δ = 6.16 (1H, d, J = 9.3 Hz, H-3), 6.73 (1H, s, H-8), 7.08 (1H, s, H-5), 7.85 (1H, d, J = 9.3 Hz, H-4).

**Cichoriin (2):** White powder; UV λ<sub>max</sub> (MeOH, nm): 226, 251, 287, 340; ¹H-NMR
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(CD$_3$OD, 500 MHz): $\delta$ = 3.42-3.55 (4H, m, H-2$'$-5$'$), 3.72 (1H, dd, J = 12.2 Hz, H-6$'$b), 4.98 (1H, d, J = 7.3 Hz, H-1$'$), 6.29 (1H, d, J = 9.3 Hz, H-3), 7.05 (1H, s, H-8), 7.21 (1H, s, H-5), 7.83 (1H, d, J = 9.3 Hz, H-4).

Luteolin (3): Pale yellow crystals; UV $\lambda_{\max}$ (nm): (MeOH) 251, 268, 348, (MeOH+NaOMe) 274, 298, 370, (MeOH+AlCl$_3$) 265, 276, 381, (MeOH+NaOAc) 262, 380, (MeOH+NaOAc+AlCl$_3$) 261, 370, (MeOH+NaOAc+H$_3$BO$_3$) 259, 380, (MeOH+NaOAc+H$_2$O) 261, 370; $^1$H-NMR (acetone-d$_6$, 500 MHz): $\delta$ = 6.32 (1H, d, J = 1.9 Hz, H-6), 6.51 (1H, d, J = 1.9 Hz, H-8), 6.61 (1H, s, H-3), 6.92 (1H, d, J = 8.4 Hz, H-5$'$), 7.43 (1H, dd, J = 1.9, 8.4 Hz, H-6$'$), 7.48 (1H, d, J = 1.9 Hz, H-2$'$).

Gossypetin (5): Yellow powder; UV $\lambda_{\max}$ (MeOH, nm): 285, 379, (MeOH+NaOMe) 283, 414, (MeOH+HCl) 279, 428, (MeOH+AlCl$_3$) 274, 298, 370, 399, 401, (MeOH+AlCl$_3$+HCl) 274, 298, 370, 381, (MeOH+NaOAc) 262, 380, (MeOH+NaOAc+AlCl$_3$) 261, 370; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 2.01 (3H, m, H-2a and H-6), 2.10 (1H, dd, J = 7.3, 14.2 Hz, H-2b), 4.10 (1H, d, J = 3.0, 7.8 Hz, H-4), 5.26 (2H, m, H-3 and H-5), 6.28 and 6.30 (1H each, d, J = 15.6 Hz, H-8$'$, H-8$''$), 6.69 and 6.70 (1H each, d, J = 8.3 Hz, H-5$'$, H-5$''$), 6.86 and 6.87 (1H each, dd, J = 1.9, 8.3 Hz, H-2$'$, H-2$''$), 7.45 (1H, d, J = 16.1 Hz, H-7$'$).

Chlorogenic acid methyl ester (9): White powder; UV $\lambda_{\max}$ (MeOH, nm): 218, 232, 244, 299, 328, 386; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 3.62 (1H, d, J = 1.9 Hz, H-6$'$a), 3.89 (3H, s, OMe), 6.41 (1H, d, J = 15.6 Hz, H-8), 6.92 (1H, d, J = 8.3 Hz, H-5$'$), 7.05 (1H, d, J = 1.9 Hz, H-8), 7.21 (1H, d, J = 15.6 Hz, H-7$'$), 7.43 (1H, s, H-3), 7.48 (1H, d, J = 16.1 Hz, H-7$'$).

Caffeic acid (7): White powder; UV $\lambda_{\max}$ (MeOH, nm): 216, 242, 295, 324, 328; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 6.42 (1H, d, J = 16.1 Hz, H-8), 6.77 (1H, d, J = 7.8 Hz, H-5), 6.95 (1H, dd, J = 1.9, 7.8 Hz, H-6), 7.07 (1H, d, J = 1.9 Hz, H-2), 7.62 (1H, d, J = 16.1 Hz, H-7); $^{13}$C-NMR (CD$_3$OD, 100 MHz, signal assignment by HSQC and HMBC experiments): 114.1 (C-2), 115.0 (C-8), 115.2 (C-5), 121.7 (C-6), 127.2 (C-1), 145.5 (C-3), 145.7 (C-7), 148.5 (C-4), 167.8 (C-9).

Ferulic acid (8): White powder; UV $\lambda_{\max}$ (MeOH, nm): 214, 232, 299, 328, 385; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 3.89 (3H, s, OMe), 6.41 (1H, d, J = 15.6 Hz, H-8), 6.80 (1H, d, J = 8.3 Hz, H-5), 7.09 (1H, dd, J = 1.9, 8.3 Hz, H-6), 7.21 (1H, brs, H-2), 7.69 (1H, d, J = 15.6 Hz, H-7); $^{13}$C-NMR (CD$_3$OD, 100 MHz, signal assignment by HSQC and HMBC experiments): 55.2 (OMe), 110.5 (C-2), 115.0 (C-8), 115.2 (C-5), 122.8 (C-6), 127.2 (C-1), 148.5 (C-3), 145.0 (C-7), 149.0 (C-4), 167.8 (C-9).

3,5-Di-O-caffeoylquinic acid (10): White powder; UV $\lambda_{\max}$ (MeOH, nm): 218, 235, 244, 299, 328; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 1.94 (3H, m, H-2a and H-6), 2.10 (1H, dd, J = 7.3, 14.2 Hz, H-2b), 4.10 (1H, dd, J = 3.0, 7.8 Hz, H-4), 5.26 (2H, m, H-3 and H-5), 6.28 and 6.30 (1H each, d, J = 15.6 Hz, H-8$'$, H-8$''$), 6.68 and 6.69 (1H each, d, J = 8.3 Hz, H-5$'$, H-5$''$), 6.86 and 6.87 (1H each, dd, J = 1.9, 8.3 Hz, H-2$'$, H-2$''$), 7.51 and 7.58 (1H each, d, J = 15.6 Hz, H-7$'$, H-7$''$).

p-Coumaric acid (6): White powder; UV $\lambda_{\max}$ (MeOH, nm): 218, 235, 244, 299, 328; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 6.42 (1H, d, J = 15.7 Hz, H-7).

3,5-Di-O-caffeoylquinic acid (10): White powder; UV $\lambda_{\max}$ (MeOH, nm): 218, 235, 244, 299, 328; $^1$H-NMR (CD$_3$OD, 500 MHz): $\delta$ = 1.94 (3H, m, H-2a and H-6), 2.10 (1H, dd, J = 7.3, 14.2 Hz, H-2b), 4.10 (1H, dd, J = 3.0, 7.8 Hz, H-4), 5.26 (2H, m, H-3 and H-5), 6.28 and 6.30 (1H each, d, J = 15.6 Hz, H-8$'$, H-8$''$), 6.68 and 6.69 (1H each, d, J = 8.3 Hz, H-5$'$, H-5$''$), 6.86 and 6.87 (1H each, dd, J = 1.9, 8.3 Hz, H-2$'$, H-2$''$), 7.51 and 7.58 (1H each, d, J = 15.6 Hz, H-7$'$, H-7$''$).
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MHz, signal assignment by HSQC and HMBC experiments): δ = 36.5 (C-2), 37.4 (C-6), 70.9 (C-4), 72.3 (C-3), 72.7 (C-5), 74.6 (C-1), 115.2 (C-8', C-8''), 115.7 and 115.8 (C-2', C-2''), 116.8 (C-5', C-5''), 123.1 and 123.2 (C-6', C-6''), 128.0 (C-1', C-1''), 145.8 (C-3', C-3''), 146.9 and 147.1 (C-7', C-7''), 148.9 and 149.3 (C-4', C-4''), 168.6 (C-9', C-9''), 175.4 (C-7).

3,5-Di-O-caffeoylquinic acid methyl ester (11): White powder; UV λ_{max} (MeOH, nm): 219, 233 sh, 244, 299 sh, 331; ESI-MS (negative): m/z 529.15 [M-1]; \( ^1H\)-NMR (CD\(_3\)OD, 500 MHz): δ = 2.15 (1H, dd, J = 6.3, 14.2 Hz, H-2a), 2.20 (2H, m, H-6), 2.31 (1H, dd, J = 3.4, 14.2 Hz, H-2b), 3.53 (3H, s, OMe), 3.98 (1H, dd, J = 2.9, 7.8 Hz, H-4), 5.32 (1H, m, H-5), 5.40 (1H, m, H-3), 6.22 and 6.34 (1H each, d, J = 15.6 Hz, H-8', -8''), 6.78 and 6.79 ( 1H each, d, J = 8.3 Hz, H-5', -5''), 6.97 (2H, brd, J = 8.3 Hz, H-6', -6''), 7.06 and 7.07 ( 1H each, d, J = 1.9 Hz, H-2', -2''), 7.55 and 7.62 ( 1H each, d, J = 15.6 Hz, H-7', -7'').

Results and Discussion

Dietary phytochemicals constitute a relevant research area of nutrition and health. The future of this area depends on the identification of active molecules within foods and plants and on an increased understanding of how the use of such molecules might play a role in disease prevention and therapy (25).

Dandelion (\textit{Taraxacum} species) contains a wide array of phytochemicals whose biological activities are actively being explored in various areas of human health. In particular, emerging evidence suggests that dandelion and its constituents have antioxidant and anti-inflammatory activities that result in diverse biological effects (1). Bitter substances are known for their stimulation of the digestion, while phenolic compounds are accounted for the anti-inflammatory and antioxidative activity of plant extracts. Therefore, focus was set on the elucidation of such pharmacologically important compounds in dandelion plants in the past decades (1, 3).

The therapeutic actions of \textit{Taraxacum} species have partially been ascribed to their bitter principles, more precisely to some sesquiterpenes. Other constituents isolated from dandelion include various triterpenes and phytosterols, phenolic compounds, and sugars, among others, found in the organs of the plant (1).

To wit, in spite of all the researches carried out, less than 1% of all the species identified so far (over 2500) have been studied (including \textit{Taraxacum officinale}, \textit{Taraxacum coreanum}, \textit{Taraxacum laevigatum}, \textit{Taraxacum mongolicum} and \textit{Taraxacum platycarpum}). This is an indication of the little knowledge that we have about this genus so far (3).

\textit{Taraxacum bessarabicum} (Hornem.) Hand.-Mazz. subsp. \textit{bessarabicum} (Hornem.) Hand.-Mazz. is a perennial herbaceous plant. The isolation of sesquiterpene lactones and two phenolics has previously been reported from the roots of \textit{Taraxacum bessarabicum} (11). Compared to roots, dandelion aerial parts are characterized by higher polyphenol contents (1). In the current study, the aerial parts of \textit{T. bessarabicum} subsp. \textit{bessarabicum} collected from Erzincan, East Anatolia were investigated to elucidate their secondary-metabolite profile. Two coumarins [esculetin (1), cichoriin (2)], three flavonoids [ luteolin (3), luteolin 7-O-β-D-glucoside (4), gossypetin (5)] and six phenolic acids and their derivatives [ p-coumaric acid (6), caffeic acid (7), ferulic acid (8), chlorogenic acid methyl ester (9), 3,5-di-O-caffeoylquinic acid (10), 3,5-di-O-caffeoylquinic acid methyl ester (11)] (Figure. 1) have been isolated from the EtOAc and CHCl\(_3\) fractions of the MeOH extract from the aerial parts of \textit{Taraxacum bessarabicum} (Hornem.) Hand.-Mazz. subsp. \textit{bessarabicum} (Hornem.). Column chromatography and preparative thin layer chromatography were used for separation of these compounds. Their structures were established conclusively by UV, ESI-MS, 1-D and 2-D NMR spectra analyses and comparison with literature data (13, 16-24). The results demonstrate that this is the first report of the isolated compounds from \textit{T. bessarabicum} subsp. \textit{bessarabicum}. Compound 5 is new for the genus \textit{Taraxacum}.

This represents the first record in the genus \textit{Taraxacum} of flavonol with extra 8-hydroxyl substituent.
Figure 1. Structures of compounds 1-11 isolated from *Taraxacum bessarabicum* subsp. *bessarabicum*.
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Acknowledgements

This work was supported by Scientific Research Projects Coordination Unit of Istanbul University (Project number: 5125, UDP-25376).

References

(1) Schütz K, Carle R and Schieber A. Taraxacum- a review on its phytochemical and pharmacological profile. J. Ethnopharmacol. (2006) 107: 313-23.
(2) Van Soest JL. Taraxacum Wiggers. In: Davis PH. (ed.) Flora of Turkey and the East Aegean Islands. Edinburgh University Press, Edinburgh (1975) 5: 788-812.
(3) Martinez M, Poirrier P, Chamy R, Prüfer D, Schulze-Gronover C, Jorquera L and Ruiz G. Taraxacum officinale and related species—an ethnopharmacological review and its potential as a commercial medicinal plant. J. Ethnopharmacol. (2015) 169: 244–62.
(4) Güner A. Taraxacum Wiggers. In: Güner A, Ozhatay N, Ekim T, Baser KHC. (eds.) Flora of Turkey and the East Aegean Islands. Edinburgh University Press, Edinburgh (2000) (Suppl. 2) 11: 170.
(5) Rajaei P and Mohamadi N. Ethnobotanical Study of Medicinal Plants of Hezar Mountain Allocated in South East of Iran. Iran. J. Pharm. Res. (2012) 11: 1153-67.
(6) Blumenthal M, Busse WR, Goldberg A, Gruenwald J, Hall T, Riggins CW and Rister RS (eds.) The Complete German Commission E Monographs. Therapeutic Guide to Herbal Medicines. American Botanical Council/Integrative Medicine Communications, Austin, Texas/Boston (1998) 118–20.
(7) Jeon HJ, Kang HJ, Jung HJ, Kang YS, Lim CJ, Kim YM and Park EH. Anti-inflammatory activity of Taraxacum officinale. J. Ethnopharmacol. (2008) 115: 82–8.
(8) Sigstедt SC, Hooten CI, Callewært MC, Jenkins AR, Romero AE, Pullin MJ, Kornienko A, Lowrey TK, Slambrouck SV and Steelant WFA. Evaluation of aqueous extracts of Taraxacum officinale on growth and invasion of breast and prostate cancer cells. Int. J. Oncol. (2008) 32: 1085–90.
(9) Altundag E and Ozturk M. Ethnomedical studies on the plant resources of East Anatolia,Turkey. Proc. Soc. Behav. Sci. (2011)19: 756–77.
(10) Shi S, Zhang Y, Huang K, Liu S and Zhao Y. Application of preparative high-speed counter-current chromatography for separation and purification of lignans from Taraxacum mongolicum. Food Chem. (2008) 108: 402–06.
(11) Kisiel W and Michalska K. Matricarin-type guaianolides from Taraxacum bessarabicum and their chemotaxonomic significance. Biochem. Syst. Ecol. (2006) 34: 356-59.
(12) Shi SY, Zhou Q, Peng H, Zhou CX, Hu MH, Tao QF, Hao XJ, Stöckigt J and Zhao Y. Four new constituents from Taraxacum mongolicum. Chin. Chem. Lett. (2007) 18: 1367–70.
(13) Williams CA, Goldstone F and Greenham J. Flavonoids, cinnamic acids and coumarins from the different tissues and medicinal preparations of Taraxacum officinale. Phytochemistry (1996) 42: 121-27.
(14) Leu Y, Shi L and Damu AG. Chemical constituents of Taraxacum formosanum. Chem. Pharm. Bull. (2003) 51: 599-601.
(15) Michalska K, Marciniuk J and Kisiel W. Sesquiterpenoids and phenolics from Taraxacum udum. Fitoterapia (2010) 81: 434-36.
(16) Schütz K, Kammerer DR, Carle R and Schieber A. Characterization of phenolic acids and flavonoids in dandelion (Taraxacum officinale Web. ex Wigg.) root and herb by high-performance liquid chromatography/electrospray ionization mass spectrometry. Rapid Commun. Mass Spectrom. (2005) 19: 179-86.
(17) Chen HJ, Inbaraj BS and Chen BJ. Determination of phenolic acids and flavonoids in Taraxacum formosanum Kitam by liquid chromatography-tandem mass spectrometry coupled with a post-column derivatization technique. Int. J. Mol. Sci. (2012) 13: 260-85.
(18) Shi SY, Zhao Y, Zhou E, Zhang Y, Jiang X and Huang K. Identification of antioxidants from Taraxacum mongolicum by high performance liquid chromatography-diode array deduction-radical scavenging deduction-electrospray ionization mass spectrometry and nuclear magnetic resonance experiments. J. Chromatogr. A (2008) 1209: 145-52.
(19) Mabry TJ, Markham KR and Thomas MB. The Systematic Identification of Flavonoids. New York, Heidelberg, Berlin (1970) 35-273.
(20) Pauli GF, Poetsch F and Nahristed A. Structure assignment of natural quinic acid derivatives using proton nuclear magnetic resonance techniques. Phytochem. Anal. (1998) 9: 177-85.
(21) Yue JM, Zhao QS, Lin ZW and Sun HD. Phenolic compounds Erigeron brevicaesp (Compositae). Acta Bot. Sin. (2000) 3: 311-15.
(22) Lee S, Park Y, Moon BH, Lee E, Hong S and Lim Y. Substitution effect of hydroxyl groups on the H and 13C chemical shifts in hydroxyflavonoids. Bull. Korean Chem. Soc. (2008) 29: 1597-99.
(23) Harborne JB. Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis. 3rd ed. Chapman and Hall Co., New York (1998) 1-302.
(24) Gawronska-Grzywacs M and Krzaczek T. Flavonoids and coumarins from Hieracium pilosella L. Acta Soc. Bot. Pol. (2009) 78: 189–95.
(25) Gonzalez-Castejon M, Visioli F and Rodriguez-Casado A. Diverse biological activities of dandelion. Nutr. Rev. (2012) 70: 534–47.

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