Epoxylignans from the seeds of *Centaurea cyanus* (Asteraceae)

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1. Subject and source

Centaurea cyanus L. (family: Asteraceae *alt*. Compositae), well known as "cornflower" or "bachelor's button", is a flowering weed endemic to Iran, Iraq, Turkey and Pakistan in Asia, and Albania, Bulgaria, Greece, Italy and Yugoslavia in Europe, and also cultivated and naturalised in many other countries of the world (GRIN database, 2003). Seeds of *C. cyanus* L. (catalogue no. 990417196) were purchased from B & T World Seeds sarl, Pauguignan, 34210 Olonzac, France, and a voucher specimen (PHSH0002) has been kept in the Plant and Soil Science Department, University of Aberdeen, UK.

2. Previous work

A variety of plant secondary metabolites including anthocyanins, flavonoids and their glycosides, phenolcarboxylic acids, polyacetylenes, sesquiterpenes, coumarins

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Table 1
^{1}H NMR (coupling constant J in Hz in parentheses) and ^{13}C NMR data of lignans **1** and **2**

Position	Chemical shifts δ in ppm			
	^{1}H		^{13}C	
	1[a]	**2**[b]	**1**[a]	**2**[b]
1			135.7	130.9
2	6.90 d (1.8)	6.92 d (1.8)	110.7	112.8
3			149.0	148.8
3-OMe	3.83 s	3.80 s	56.4	55.9
4			147.1	147.4
5	6.89 d (8.2)	6.72[c]	116.0	115.7
6	6.74 dd (8.2, 1.8)	6.72[c]	119.8	121.7
7	4.74 d (6.9)	4.80 s	84.1	85.8
8	2.37 m		54.0	83.4
9	3.82[c] 3.64 dd (11.6)	3.76 d (11.6), 3.56 d (11.6)	60.5	64.6
1′			137.2	133.4
2′	6.77 d (1.8)	6.77 d (1.8)	114.4	113.8
3′			150.9	149.2
3′-OMe	3.84 s	3.81	56.8	56.1
4′			146.4	146.0
5′	7.08 d (8.2)	6.68 d (8.0)	118.4	116.4
6′	6.73 dd (1.8, 8.2)	6.61 dd (8.0, 1.8)	122.3	122.5
7′	2.98 dd (13.5, 4.7), 2.54 dd (13.5, 11.3)	3.05 dd (12.8, 3.8), 2.43 t (12.8, 12.4)	33.7	35.2
8′	2.73 m	2.54 m	43.7	52.0
9′	3.96 dd (8.0, 5.2), 3.70 dd (8.0, 6.6)	4.03 dd (8.0, 5.2), 3.62 dd (8.0, 6.6)	73.5	72.1
1″	4.84[c]	–	103.1	
2″	3.47[c]	–	75.0	
3″	3.44[c]	–	77.9	
4″	3.30[c]	–	71.4	
5″	3.38[c]	–	78.2	
6″	3.88[c] 3.68[c]	–	62.6	

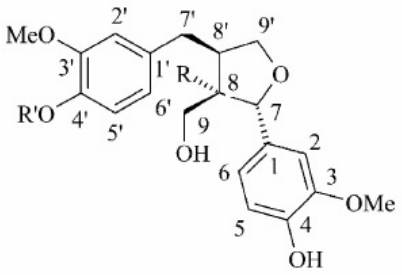
Spectra obtained in CD_3OD.
[a] ^{1}H NMR (600 MHz) and ^{13}C NMR (150 MHz).
[b] ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz).
[c] Overlapped signals, assignment was confirmed from TOCSY, COSY and HSQC correlations.

and indole alkaloids have previously been reported from *C. cyanus* (Dictionary of Natural Products, 2001; Hegnauer, 1964; Phytochemical and Ethnobotanical database, 2003; Sarker et al., 2001).

3. Present study

Ground seeds (100 g) of *C. cyanus* were Soxhlet-extracted, successively, with hexane, dichloromethane and MeOH. All these extracts were separately

concentrated using rotary evaporator at a maximum temperature of 40 °C. The MeOH extract was fractionated on a Sep-Pak C_{18} (10 g) cartridge, using 30%, 40%, 60%, 80% and 100% MeOH–water mixture (200 ml each) as eluent. Preparative RP-HPLC (Luna C_{18} column 10 μm, 250 × 21.2 mm, eluted with a linear gradient—water : ACN = 80 : 20–40 : 60 over 50 min followed by 60% ACN for 10 min, 20 ml/min, monitored by photo-diode-array detector) of the Sep-Pak fraction eluted with 40% aqueous MeOH yielded epoxylignans lariciresinol 4′-*O*-β-D-glucopyranoside (**1**, 4.7 mg, t_R = 17.1 min) (Dellagreca et al., 1993) and berchemol (**2**, 10.3 mg, t_R = 22.2 min) (Sakurai et al., 1989). Compounds **1** and **2** were identified unequivocally by UV, ESIMS, HRMS, ^{1}H and ^{13}C NMR and a series of 2D NMR analyses. Unambiguous and complete assignment of all ^{1}H and ^{13}C NMR signals for **1** and **2**, on the basis of ^{13}C DEPT135, COSY, TOCSY, HMQC, HMBC and NOESY experiments, is presented here (Table 1).



1 R = H R′ = Glucosyl

2 R = OH R′ = H

4. Chemotaxonomic significance

To our knowledge, this is the first report on the occurrence of epoxylignans, berchemol (**1**) and lariciresinol 4-*O*-β-D-glucopyranoside (**2**) in the seeds of *C. cyanus*. While the genus *Centaurea* is known to produce predominantly dibenzylbutyrolactone-type lignans (Middleton et al., 2003; Cooper et al., 2002; Ferguson et al., 2002; Ribeiro et al., 2002), epoxylignans, e.g. olivil, lappaol A, pinoresinol and lirioresinol also occur in a few *Centaurea* species. However, lariciresinol-type epoxylignans **1** and **2** have never been reported from any *Centaurea* species. Within the family Asteraceae, this type of lignans has previously been reported from *Vladimiria souliei* (Tan et al., 1990). Lariciresinol 4′-*O*-β-D-glucopyranoside (**1**) also occurs in a few species of the genera *Arum* (Family: Araceae), *Osmanthus* and *Syringa* (Family: Oleaceae), and *Rhodiola* (Family: Crassulaceae) (Dictionary of Natural Products, 2001). Apart from *C. cyanus* and *Vladimiria souliei* of the Asteraceae, the distribution of berchemol (**2**) appears to be limited to *Berchemia racimosa* of the family Rhamnaceae (Sakurai et al., 1989).

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References

Cooper, G., Laird, A., Nahar, L., Sarker, S.D., 2002. Biochem. Syst. Ecol. 30, 65–67.

Dellagreca, M., Molinaro, A., Monaco, P., Previtera, L., 1993. Heterocycles 36, 2081–2086.

Dictionary of Natural Products (DNP) on CD-ROM, 2001. Chapman and Hall, Boca Raton, Florida.

Ferguson, C.A., Nahar, L., Finnie, D., Kumarasamy, Y., Reid, R., Mir-Babayev, N.F., Sarker, S.D., 2002. Biochem. Syst. Ecol. 31, 303–305.

GRIN database, 2003. USDA, ARS, National Genetic Resources Program. Germplasm Resources Information Network (GRIN), National Germplasm Resources Laboratory, Beltsville, Maryland, USA. Available from http/www.ars-grin.gov/cgi-bin/npgs/html/tax_search.pl?*Centaurea+cyanus*.

Hegnauer, R., 1964. Chemotaxonomie der Pflanzen, vol. III, p. 502, Basel Birkhauser Verlag.

Middleton, M., Cox, P.J., Jaspars, M., Kumarasamy, Y., Nahar, L., Reid, R., Sarker, S.D., 2003. Biochem. Syst. Ecol. 31, 653–656.

Phytochemical and Ethnobotanical database, 2003. USDA-ARS-NGRL, Beltsville Agricultural Research Center, Beltsville, Maryland, USA. Available from http://www.ars-grin/cgi-bib/duke/pharmacy2.pl.

Ribeiro, N.L., Nahar, L., Kumarasamy, Y., Mir-Babayev, N., Sarker, S.D., 2002. Biochem. Syst. Ecol. 30, 1097–1100.

Sakurai, N., Nagashima, S., Kawai, K., Inoue, T., 1989. Chem. Pharm. Bull. 37, 3311–3315.

Sarker, S.D., Laird, A., Nahar, L., Kumarasamy, Y., Jaspars, M., 2001. Phytochemistry 57, 1273–1276.

Tan, R.X., Jakupovic, J., Jia, Z.J., 1990. Planta Med. 56, 475–477.