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CONYZATIN—A NEW FLAVONE FROM *CONYZA STRICTA**

SHEELA TANDON and R. P. RASTOGI

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Central Drug Research Institute, Lucknow, India

Key Word Index—*Conyza stricta*; Compositae; 5,7-dihydroxy-3,8,4′-trimethoxyflavone; 5,7-dihydroxy-3,8,3′,4′,5′-pentamethoxy flavone.

Conyza stricta Willd. is a small herb found distributed over subtropical Himalayas and Western Ghats up to 1525m. Chemical examination of *C. filaginoides* has been reported to yield triacontanes, α-spinasterol and β-amyrin [1]. A bitter substance, conyzin, from the leaves of *C. discoridis* [2] and a furanoditerpene, hautriwaic acid, from the *C. ivaefolia* [3] have been reported. Chemical studies on *C. bonariensis* [4] have been reported to yield first naturally occurring cumelene. The alcoholic extract of *C. stricta* exhibited antiamphetamine activity [5].

The ethyl acetate soluble portion of the plant extract was subjected to Si gel chromatography which resulted in the isolation of two flavones designated as E and F, the latter substance being new, has been named as conyzatin. Substance E, $C_{18}H_{16}O_7$, mp 173°, showed colour reactions of flavonoids. Its UV showed absorptions at 277, 312 and 358 nm and IR indicated the presence of a chelated carbonyl (1650 cm^{-1}) along with hydroxy and aromatic bands in the usual regions. PMR spectrum displayed signals for three methoxyls at 3.9 ppm, one proton singlet at 6.4 ppm and two *ortho* coupled doublets of two protons at 7.0 and 8.05 ppm (each $J = 9$ Hz). Substance E yielded a diacetate, a dimethyl ether with DMS and a monomethyl ether with CH_2N_2, which was characterised as flindulatin [6]. Substance E was identified as 5,7-dihydroxy-3,8,4′-trimethoxyflavone [7] by comparison of data with an authentic sample.

Conyzatin, $C_{20}H_{20}O_9$ (M^+ 404) was indicated to be a C-3 oxygenated flavonoid by its colour reactions and similarity of the spectral patterns with that of substance E. It showed UV maxima at 276, 314, 362, 402 nm and IR bands for hydroxyl (3350), chelated carbonyl (1650) and aromatic moiety (1600, 1575, 1500 cm^{-1}). The PMR spectrum displayed signals for five methoxyls at 3.8–3.9, a one proton and a two proton singlets at 6.25 and 7.45 ppm respectively.

The formation of conyzatin diacetate, $C_{24}H_{24}O_{11}$, indicated the presence of two hydroxyl groups which were confirmed as phenolic by the formation of dimethyl ether ($C_{22}H_{24}O_9$) with DMS and monomethyl ether ($C_{21}H_{22}O_9$) with CH_2N_2. The position of these hydroxyls was assigned on the basis of UV spectral data.

The UV spectrum of conyzatin remained unchanged on addition of NaOAc–H_3BO_3 indicating the absence of *o*-dihydroxyls in the molecule. When UV was recorded with $AlCl_3$ four major bands (IIb, IIa, Ib and Ia) were observed at 286, 317, 355, 400 and 421 (*sh*) which persisted on addition of conc HCl. The observed 38 nm bathochromic shift of the band I to Ia suggested that one of the hydroxyls was located at C-5, since in C-3 hydroxyflavonoids, the magnitude of the shift of band I to Ia is of the order of 60 nm. This placement was in agreement with the formation of monomethyl ether with CH_2N_2 because of the inertness of C-5 hydroxyl group to this reagent. The 8 nm bathochromic shift of the band II on addition of NaOAc indicated that second hydroxyl group must be located at C-7.

The mass fragmentation pattern of the conyzatin was not very characteristic due to highly substituted pattern in the rings A and B, but the intense peaks at *m/e* 403 (M^+-1) and *m/e* 389 (M^+-M3) indicated the presence of C-3 OMe and C-6/C-8 OMe groups respectively. The presence of two proton singlet at 7.4 ppm was in the range characteristic for C-2′6′ protons of flavonoids having C-3′,4′,5′ oxygenation pattern which indicated that ring B was symmetrically substituted by methoxy group at C-3′,4′ and 5′. The remaining two methoxyls and one proton singlet at 6.25 ppm were left to be assigned at either of C-3, C-6 or C-8 positions. The appearance of conyzatin as purple spot under UV indicated the presence of OH at C-5 and OMe at C_3; the latter grouping was further supported by the colour reaction with Mg/HCl. Further since C-8 proton in 5,6,7-trimethoxy flavonoids is reported [8] to appear at 6.77 ppm. it was therefore, assigned to C-6 which was substantiated by similar chemical shift of C-6 proton in 5,7,8-trimethoxyflavonoids. The C-3 and C-8 positions were consequently left for the assignment of remaining two methoxyls. The presence of 5,7-dihydroxy grouping in the molecule was further supported by the downfield shift (0.8 ppm) of C-6 proton on acetylation of conyzatin. On the basis of the above data, the structure of conyzatin was assigned as 5,7-dihydroxy-3,8,3′,4′,5′-pentamethoxyflavone.

The proposed structure was confirmed by comparing the data of conyzatin dimethyl ether with an authentic sample of hibiscetin heptamethyl ether [9].

EXPERIMENTAL

The reported mps are uncorr. The PMR spectra were recorded in $CDCl_3$ unless stated otherwise, with TMS as internal standard. The R_f values relate to Si gel plates using

*CDRI Communication No. 2307 *Conyza stricta* was collected from Dehradun, U.P. and identified by Mr. B. N. Mehrotra. A voucher specimen No. 3276 has been preserved in the herbarium of the Institute.

ceric sulphate as spray reagent. The EtOH extractive of the dry plant material (2 kg) was macerated with EtOAc and the soluble fraction (92 g) was chromatographed over Si gel (1 kg). Fifty fractions (500 ml each) were collected using hexane containing increasing amounts of Et_2O. The hexane–Et_2O (1:1) eluates (fraction 22–24, 3.686 g) yielded substance E (1.068 g) after purification by PLC. Subsequent fractions 35–47 of the same eluant (2.786 g) on crystallization with MeOH yielded substance F (500 mg).

Substance E (5,7-dihydroxy-3-8,4'-trimethoxyflavone). Yellow needles from dil. MeOH mp 172–73°, R_f 0.35 (hexane–Et_2O 1:1). It produced magenta colour with Mg/HCl and an intense yellow with ammonia. UV (MeOH): 277, 312, 358 nm (log ε 4.37, 3.83, 3.7); MeOH/$AlCl_3$: 285, 311, 352, 412 nm; MeOH/NaOAc: 285, 307, 390 nm: MeOH/NaOMe: 285, 305, 415 nm. PMR (acetone-d_6) ppm: 3.9 (9H, *s*, 3OMe), 6.3 (1H, *s*, C6), 7.1 (2H, *d*, J = 9 Hz, C3'5'), 8.1 (2H, *d*, J = 9 Hz, C2'6'). MS: *m/e* 344 (M^+). Found: C, 62.2; H, 4.2 $C_{18}H_{16}O_7$ requires C, 62.0; H, 4.2%. The diacetate ($C_{22}H_{20}O_9$) crystallized from $CHCl_3$–MeOH, mp 160–62°. UV (MeOH): 265 and 340 nm. PMR ppm: 2.35, 3.45 (3H each, *s*, OAc), 3.84 (3H, *s*, OMe), 3.9 (6H, *s*, 2OMe), 6.8 (1H, *s*, C6), 7.12 (2H, *d*, J = 9 Hz, C3'.5'), 8.12 (2H, *d*, 9 Hz, C2',6'). The monomethylether ($C_{19}H_{18}O_7$) crystallized from dil. EtOH, mp 161°. PMR ppm: 3.9 (12H, *s*, 4OMe), 6.4 (1H, *s*, C6), 7.0 (2H, *s*, J = 9 Hz, C3',5'), 8.05 (2H, *d*, J = 9 Hz, C2',6'). The dimethyl ether ($C_{20}H_{20}O_7$) crystallized from dil. MeOH, mp 156°. UV (MeOH): 272, 310, 350 nm. PMR ppm: 3.9 (15H, *s*, 5OMe), 6.4 (1H, *s*, C6), 7.0 (2H, *d*, J = 9 Hz, C3',5'), 8.05 (2H, *d*, J = 9 Hz, C2',6').

Substance F (conyzatin). Yellow needles from MeOH, mp 209°. With Mg/HCl it gave a magenta colour and an intense yellow spot with NH_3. R_f 0.4 (hexane–Et_2O 1:3). UV (MeOH): 276, 314, 362, 402 nm (log *e* 4.42, 4.25, 3.90, 3.98); MeOH/$AlCl_3$: 286, 317, 355, 400, 421 nm: MeOH/NaOAc: 287, 305, 414, MeOH/NaOMe: 288, 305, 416 nm. PMR (DMSO-d_6) ppm: 3.83 (3H, *s*, OMe), 3.9 (12H, *s*, 4OMe), 6.25 (1H, *s*, C6). 7.4 (2H, *s*, C2',6'), MS: *m/e* (M^+) 404. Found: C, 58.35: H, 6.1 $C_{20}H_{20}O_9$ requires C, 58.30; H, 6.05%. The diacetate was obtained as amorphous powder from hexane–C_6H_6, mp 152°. PMR (CCl_4) ppm: 2.23, 2.29, (3H each, *s*, OAc), 3.8 (12H, *s*, 4OMe), 3.9 (3H, *s*, OMe), 6.45 (1H, *s*, C6), 7.3 (2H, *s*, C2',6'). PMR (DMSO-d_6) ppm: 2.3 (6H, *br s*, 2OAc), 3.75–3.85 (12H, *br s*, 4OMe), 4.0 (3H, *s*, OMe), 7.05 (1H, *s*, C6), 7.38 (1H, *s*, C2'), 7.6 (1H, *s*, C6'). Found: C, 58.1; H, 4.8 $C_{24}H_{24}O_{11}$ requires C, 59.12, H, 4.9%. The monomethyl ether crystallised from MeOH, mp 170°. UV (MeOH): 275, 308, 340, 362 nm; MeOH/$AlCl_3$: 285, 320, 356, 400 nm. PMR (acetone-d_6) ppm: 3.8 (3H, *s*, OMe), 3.85 (3H, *s*, OMe), 3.86 (9H, *s*, 3OMe), 3.91 (3H, *s*, OMe), 6.4 (1H, *s*, C6), 7.46 (2H, *s*, C2',6'). MS: *m/e* 418 (M^+). The dimethyl ether crystallized from MeOH, mp 192°. UV (MeOH): 277, 308, 356 nm; UV ($AlCl_3$, NaOAc): no change PMR (DMSO-d_6) ppm: 3.78 (3H, *s*, OMe), 3.8 (3H, *s*, OMe), 3.85 (3H, *s*, OMe), 3.88 (9H, *s*, 3OMe), 3.98 (3H, *s*, OMe), 6.3 (1H, *s*, C6), 7.42 (2H, *s*, C2',6'). MS: *m/e* 432 (M^+).

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BIFLAVONES AND FLAVONOL-*O*-GLYCOSIDES FROM *JUNIPERUS MACROPODA*

MOHAMMAD ILYAS, NAJMA ILYAS and HILDEBERT WAGNER
Institut für pharmazeutische Arzneimittellehre der Universität München, BRD

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Key Word Index—*Juniperus macropoda*; Cupressaceae; amentoflavone; hinokiflavone; isocryptomerin; quercetagetin-3-*O*-rhamnoside; quercetin-3-*O*-rhamnoside; kaempferol-3-*O*-glucoside.

Powdered and dried leaves of *Juniperus macropoda* Boiss. were extracted with acetone in a Soxhlet. The extract was dried under reduced pressure and treated with hot water. The insoluble portion was purified by column chromatography on silica gel to give three biflavones by preparative TLC. They were characterized as amentoflavone, hinokiflavone and isocryptomerin, by mp and comparison of NMR spectra of their methyl and acetyl derivatives with those of authentic samples.

The water soluble portion was extracted with butanol and separated by column chromatography followed by TLC on silica gel into three fractions A, B, and C. On hydrolysis with 10% HCl fractions A and B gave quercetagetin, mp 320–324° and quercetin, mp 315° respectively and a common sugar rhamnose (TLC), while fraction C gave kaempferol, mp 284° and glucose. The aglycones were characterized by co-chromatography on TLC and by comparing the mp's of their acetates with authentic