Phytochemistry, Vol. 21, No. 8, pp. 2145–2146, 1982.
Printed in Great Britain.

0031-9422/82/082145-02$03.00/0

3, 4, 7-TRIMETHYLCOUMARIN FROM *TRIGONELLA FOENUM-GRAECUM* STEMS

SHASHI K. KHURANA, VELAGALETY KRISHNAMOORTHY, VIRINDER S. PARMAR,* RADHIKA SANDUJA* and HARMOHINDER L. CHAWLA*

Sri Venkateswara College (University of Delhi), Dhaula Kuan, New Delhi-110021, India; *Department of Chemistry, University of Delhi, Delhi-110007, India

(*Received* 31 *December* 1981)

Key Word Index—*Trigonella foenum-graecum*; Leguminosae; stem; 3, 4, 7-trimethylcoumarin.

Abstract—A trimethylcoumarin has been isolated, for the first time from the stem of *Trigonella foenum-graecum* along with some known compounds. Based upon spectral and analytical data and comparison with a synthetic sample, it has been assigned the structure 3, 4, 7-trimethylcoumarin.

All parts of the medicinal plant *Trigonella foenum-graecum*, except the stems, have been extensively studied for their chemical components and have been shown to contain steroidal sapogenins [1, 2], carotenoids [3], flavonoids [4–6] and coumarins ([7]; Jha, H. N., Sanduja, S. K., Sanduja, R. and Parmar, V. S., personal communication). Only one compound (diosgenin) has been reported [8] from the stem; in the present study, we have examined the stem in detail. This has resulted in the isolation of 3, 4, 7-trimethylcoumarin for the first time from any natural source.

As detailed in the Experimental, eight compounds have been isolated by CC and prep. TLC of concentrates of the hot benzene and alcohol extracts of the stem portion of *T. foenum-graecum* (compounds A–D from the benzene extract and compounds E–H from the alcohol extract). From physical and spectroscopic data and also by comparison with authentic samples, seven of these compounds were found to be known products, namely sitosterol (A), 4-methyl-7-acetoxycoumarin (C), *p*-coumaric acid (D), luteolin (E), quercetin (F), diosgenin (G) and vitexin (H). The characterization of the remaining compound (B) is described.

Compound B ($C_{12}H_{12}O_2$, from its analytical data and M^+ peak at *m/z* 188 in its mass spectrum) crystallized from methanol as colourless shining needles, mp 112–113° and exhibited a blue fluorescence in UV light. It did not furnish any colour with alcoholic ferric chloride and also did not respond to any metal–acid reduction tests. It dissolved in aq. NaOH to give a deep yellow colour and also gave a yellow colour with hot conc. H_2SO_4. Its IR spectrum showed strong absorption bands at 1695 and 1610 cm^{-1}, characteristic of an α, β-unsaturated six-membered lactone ring of the coumarin type [9]. In its UV spectrum, B showed λ_{max} at 242(sh), 280 and 310 nm and did not produce any bathochromic shift with NaOAc and $AlCl_3$. The above colour reactions and absorption spectra suggest that B is a coumarin without any hydroxyl group. Its NMR spectrum ($CDCl_3$) showed the presence of three C-methyl groups as sharp singlets at δ 2.18, 2.35 and 2.40, and three aromatic protons as an *ortho*-coupled doublet at δ 7.45 (J = 9 Hz, 1H) and an ill-resolved double-doublet at δ 7.06 (2H). It is well known that the C-3 and C-4 protons appear in the ranges, δ 6.11–6.39 and δ 7.85–8.15, respectively, as *ortho* coupled doublets if both the positions are free. If one of these positions is carrying a methyl group, however, the neighbouring proton appears as a quartet. The absence of any such signals in the NMR spectrum of B indicated that both the C-3 and C-4 positions are substituted by methyl groups. The remaining methyl group is, thus, located in the aromatic ring. It has been reported [10] that the C-5 and C-7 protons of coumarins appear more downfield than the C-6 and C-8 protons. As the NMR spectrum of B showed only one aromatic proton downfield and two protons comparatively upfield, the third methyl group could, therefore, be placed at either C-5 or C-7. It has also been noted [10–12] that the C-5 methyl group appears around δ 2.60 while the C-7 methyl appears around δ 2.36. As the third methyl group in B appeared at δ 2.35, it indicated that it is present at C-7. Based upon the above analysis of its NMR spectrum, B was given the structure 3, 4, 7-trimethylcoumarin. This structure for B is fully supported by the fragmentation pattern in its mass spectrum. Intense peaks were observed at *m/z* 188 (M^+), 160 ($M^+ - CO$), 145, 130 and 115 (due to the successive losses of the three C-methyl groups). The structure is also supported by comparing the values of the benzene-induced solvent shifts of the three C-methyl groups (as given in the Experimental section) with those in the literature [12]. The above structure of B was finally proved by comparing it with a synthetic sample of 3, 4, 7-trimethylcoumarin, obtained by condensation of *m*-cresol and monomethylethyl acetoacetate in the presence of conc. H_2SO_4, in the cold. The two samples agreed very well. This is the first report of this coumarin and we

propose the name 'trigoforin' for this. We may mention that coumarins lacking oxygenated substituents are of very rare natural occurrence.

EXPERIMENTAL

The IR spectra were recorded in KBr pellets while UV spectra were recorded in methanol. The NMR spectra were obtained with TMS as int. standard. Mps were determined in H_2SO_4 bath. For CC, Si gel-G (90–200 mesh, E. Merck) was used while prep. TLC was carried out on 0.5 mm Si gel-G (type 60, E. Merck) layers.

The whole plant of *Trigonella foenum-graecum* was purchased from the local market of Delhi and identified in the Department of Botany, University of Delhi. The stem was separated, the air-dried stems (1.5 kg) were extracted first with C_6H_6 (3 × 2 l.) and then with C_2H_5OH (4 × 2 l.) in hot. The C_6H_6 extract, on removal of the solvent, yielded a greenish gummy residue which was chromatographed on a Si gel column and the column was eluted with mixtures of petrol–C_6H_6 and C_6H_6–EtOAc in varying proportions. The earlier fractions yielded A in pure form, fractions eluted with pure C_6H_6 contained a mixture of B and C while D was obtained from fractions eluted with C_6H_6–EtOAc (49:1 and 19:1). The mixture of B and C was separated by prep. TLC using C_6H_6–CH_3COCH_3 (19:1). The total EtOH extract on removal of the solvent gave a brownish green viscous mass and was chromatographed on a column of Si gel, the eluants being mixtures of C_6H_6, EtOAc and MeOH in different proportions. Compounds E–H were obtained from different fractions.

Trigoforin (B) crystallized from MeOH as colourless shining needles (0.03 g), mp 112–113°. (Found: C, 76.22, H, 6.85, $C_{12}H_{12}O_2$ requires: C, 76.59, H, 6.38%.) UV λ_{max} (nm): 242(sh), 280 and 310. IR ν_{max} (cm^{-1}): 1695, 1610, 1555, 1385, 1348, 1322, 1095, 894, 840, 763 and 735. EIMS (probe) 70 eV, *m/z* (rel. int.): 188(100), 160(89), 159(70), 145(90), 115 (36), 91(30), 77(23), 65(18), 51(30). ^1H NMR (in $CDCl_3$): δ2.18 (3H, *s*, C-3 Me), 2.35 (3H, *s*, C-7 Me), 2.40 (3H, *s*, C-4 Me), 7.06 (2H, *dd*, C-6 and C-8H), 7.45 (1H, *d*, $J = 9$ Hz, C-5H). ^1H NMR (in 1:1 $CDCl_3 + C_6H_6$): δ 1.19 (3H, *s*, C-4 Me), 1.98 (3H, *s*, C-7 Me) and 2.14 (3H, *s*, C-3 Me).

Synthesis of 3, 4, 7-*trimethylcoumarin.* To the ice-cold mixture of *m*-cresol (10 g) and monomethyl derivative of ethylacetoacetic ester (10 g), conc. H_2SO_4 (20 ml, *d* 1.84) was added slowly with continuous stirring. The solution was kept in the refrigerator overnight and then poured into ice-cold water (200 ml) with continuous stirring. The solid that separated was filtered, washed with water and crystallized from MeOH as colourless shining needles, mp 112°. Mmp with the natural material (B) remained unchanged and also both samples produced superimposable IR spectra.

REFERENCES

1. Varshney, I. P. and Sharma, S. C. (1966) *J. Indian Chem. Soc.* **43**, 564.
2. Fazli, F. R. Y. and Hardman, R. (1968) *Tropical Sci.* **10**, 66.
3. Sadana, J. C. and Ahmad, B. (1949) *J. Sci. Ind. Res.* **8B**, 52.
4. Ademska, M. and Lutomski, J. (1971) *Planta Med.* **20**, 224.
5. Wagner, H., Iyengar, M. A. and Hörhammer, L. (1973) *Phytochemistry* **12**, 2548.
6. Sood, A. R., Boutard, B., Chadenson, M., Chopin, J. and Lebreton, P. (1976) *Phytochemistry* **15**, 351.
7. Bhardwaj, D. K., Murari, R., Seshadri T. R. and Singh, R. (1977) *Indian J. Chem.* **15B**, 94.
8. Varshney, I. P. and Sood, A. R. (1971) *Indian J. Appl. Chem.* **34**, 208.
9. Dean, F. M. (1963) *Naturally Occurring Oxygen Ring Compounds*, p. 176. Butterworths, London.
10. Rowbotham, J. B. and Schaefer, T. (1973) *Can. J. Chem.* **51**, 953.
11. Chexal, K. K., Fourwather, C. and Holker, J. S. E. (1975) *J. Chem. Soc. Perkin Trans.* 1, 554.
12. Grigg, R., Knight, J. A. and Roffey, R. (1966) *Tetrahedron* **22**, 3301.