3686 *J. Org. Chem.* **1981**, *46*, 3686-3688

Synthesis of Two Aromatic Methylenedioxy-Substituted Colchicine Congeners: Elucidation of the Structure of Cornigerine as 2,3-(Methylenedioxy)-2,3-Didemethoxycolchicine

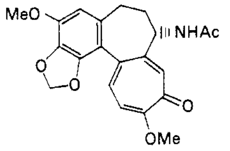
Manfred Rösner,[1] Fu-Lian Hsu, and Arnold Brossi*

Section on Medicinal Chemistry, Laboratory of Chemistry, National Institute of Arthritis, Metabolism and Digestive Diseases, National Institutes of Health, Bethesda, Maryland 20205

Received April 15, 1981

Treatment of colchicine with concentrated sulfuric acid at 80–90 °C afforded besides 2-demethylcolchicine a mixture of catecholic colchicines which could be separated and identified. Similar treatment of 1-demethyl- and 3-demethylcolchicine gave predominantly the 1,2-catechol or its 2,3 isomer. Methylenation of these catechols afforded the colchicine congeners 1 and 10. A comparison with natural cornigerine revealed that the alkaloid was identical with the 2,3-methylenedioxy compound 10 and not with 1, as formerly proposed.

Cornigerine, an alkaloid from *Colchium cornigerum* species, was isolated by Šantavý et al. in 1961[2] and reported to exhibit similar biological activities as colchicine.[3] Structure 1 for cornigerine, with a 1,2-methylenedioxy ring annulated to the aromatic ring A of colchicine replacing the two original methoxy groups, was proposed on the basis of a ^{1}H NMR analysis and by chemical degradation[4] (see structures 1 and 10). It is interesting to note that the postulated structure of cornigerine differed in the aromatic A region with respect to the methylenedioxy substitution pattern from that established for the accompanying minor alkaloids CC-20 and CC-2.[5] It was also interesting to note that Šantavý's groups concluded that the singlet at δ 6.01 marked the protons of the methylenedioxy group, contrary to reports that hindered aromatic methylenedioxy groups have unequal protons, frequently shown as doublets.[6]



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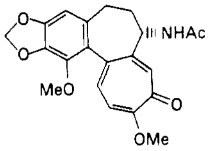
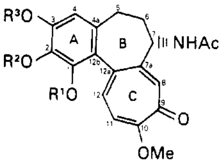


10

CORNIGERINE

These discrepancies, together with a recently accomplished selective O-demethylation of the ring-A-positioned methoxy groups of colchicine,[7,8] suggested the preparation of cornigerine by partial synthesis. Treatment of colchicine (2) with concentrated sulfuric acid at 80–90 °C afforded, besides a small amount of 2-demethylcolchicine (4), the major reaction product obtained at 60 °C,[7] a slightly more polar substance (TLC; see structures 2–9). The dark blue color shown after the plate was sprayed with an alcoholic ferric chloride solution hinted that this substance might be of catecholic nature. The same product was formed by similar treatment of 1-demethylcolchicine (3)[7,8] or 2-demethylcolchicine (4)[7,8] with concentrated sulfuric acid. It could be assumed that the sterically most hindered 2-methoxy group in colchicine was preferentially cleaved[9] and that catechol 6 was formed, probably via the intermediate monophenol 4. Catechol 6, isolated from the sulfuric acid treatment of colchicine in 10% yield, was indeed identical in its chromatographic properties with the newly formed catechol. The ^{1}H NMR spectrum of 6 showed the signals of the protons of the 3- and 10-methoxy groups as singlets at δ 3.80 and 3.84 respectively (Table I), in good agreement with earlier findings.[10-12] Acetylation of 6 afforded the diacetate 7, and further reaction of 6 or 7 with bromochloromethane, in the presence of potassium carbonate in 1-methyl-2-pyrrolidinone at 70 °C, gave the crystalline 1,2-methylenedioxy-substituted derivative 1. Although 1 could not be differentiated from natural cornigerine by TLC and mass spectroscopy, its optical rotation and IR (Figure 1) and NMR spectra (Table I), were quite different from those of the natural alkaloid, suggesting that the latter did not have the proposed structure. Particularly the protons assigned to the methylenedioxy group, showing a doublet at δ 5.91 and 6.05 (Table I), were clearly different from the singlet at δ 6.01 reported by Šantavý et al.[4] Our data suggested that natural cornigerine would be much better accomodated with structure 10.



2 $R^1 = R^2 = R^3 = CH_3$

3 $R^1 = H; R^2 = R^3 = CH_3$

4 $R^2 = H; R^1 = R^3 = CH_3$

5 $R^3 = H; R^1 = R^2 = CH_3$

6 $R^1 = R^2 = H; R^3 = CH_3$

7 $R^1 = R^2 = Ac; R^3 = CH_3$

8 $R^2 = R^3 = H; R^1 = CH_3$

9 $R^2 = R^3 = Ac; R^1 = CH_3$

The starting material for the prepartion of 10 was 3-demethylcolchicine (5), readily obtained from colchicoside by cleavage of the glycosidic linkage with phosphoric acid.[7]

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[1] Guest scientist from the Pharmaceutical Division of Hoechst AG, Frankfurt, Federal Republic of Germany.

[2] El-Hamidi, A.; Šantavý, F. *Collect. Czech. Chem. Commun.* **1962**, *27*, 2111.

[3] Šantavý, F. *Acta. Univ. Palacki. Olomouc.* **1979**, *90*, 15–44. We thank Professor Šantavý for having sent us a copy of this review article.

[4] Cross, A. D.; El-Hamidi, A.; Hrbek, Jr., J.; Šantavý, F. *Collect. Czech. Chem. Commun.* **1964**, *29*, 1187.

[5] Battersby, A. R.; Ramage, R.; Cameron, A. F.; Hannaway, C.; Šantavý, F.; *J. Chem. Soc. C* **1971**, 3514.

[6] Brossi, A.; Blount, J. F.; O'Brien, J.; Teitel, S. *J. Am. Chem. Soc.* **1971**, *93*, 6248.

[7] Rösner, M.; Capraro, H.-G.; Iorio, M. A.; Jacobson, A. E.; Atwell, L.; Brossi, A.; Williams, T. H.; Sik, R. H.; Chignell, C. F. *J. Med. Chem.* **1981**, *24*, 257.

[8] Bladé-Font, A. *Afinidad* **1979**, *36*, 329.

[9] Brossi, A.; Teitel, S. *Chem. Commun.* **1970**, 1296.

[10] Capraro, H.-G.; Brossi, A. *Helv. Chim. Acta* **1979**, *62*, 965.

[11] Hufford, C. D.; Capraro, H.-G.; Brossi, A. *Helv. Chim. Acta* **1980**, *63*, 50.

[12] Elguero, J.; Muller, R. N.; Bladé-Font, A.; Faure, R.; Vincent, E. J. *Bull. Soc. Chim. Belg.* **1980**, *89*, 193.

0022-3263/81/1946-3686$01.25/0

Table I. ^{1}H NMR Data of Colchicine and Derivatives

	chemical shift,[d] δ					
compd	*N*-$COCH_3$	1-OCH_3	2-OCH_3	3-OCH_3	10-OCH_3	$OCOCH_3$
		In Me_2SO-d_6				
3	1.84		3.71	3.80	3.84	
4	1.84	3.45		3.81	3.86	
5	1.84	3.49	3.76		3.84	
6	1.84			3.80	3.84	
8	1.83	3.42			3.84	
		In $CDCl_3$				
2	1.95	3.64	3.88[a]	3.92[a]	4.00	
7	1.95			3.86	3.96	2.16, 2.29
9	1.96	3.57			3.99	2.31, 2.33
1	1.93	5.91/6.05[b]		3.92	3.96	
10	1.96	3.77	5.96[c]		3.97	
natural cornigerine	1.96	3.77	5.96[c]		3.97	

[a] May be reversed. [b] 1,2-(OCH_2O), AB signal, apparent doublet, $J_{gem} < 2$ Hz. [c] 2,3-(OCH_2O), singlet. [d] All listed signals, except that of footnote *b*, refer to singlets.

Treatment of **5** with sulfuric acid at 80–90 °C afforded as the major reaction product the expected catechol **8**, identical by TLC with a minor component obtained in the sulfuric acid treatment of **4**. The signals of the 1- and 10-methoxy protons of **8** are shown as singlets at δ 3.42 and 3.84, respectively (Table I), in agreement with published data.[11,12]

Catechol **8** seemed, in addition, to be identical with a natural product isolated by Šantavý et al. from *Colchicum latifolium*.[13] Conversion of **8**, or its diacetate **9**, into cornigerine was accomplished by alkylation with bromo chloromethane, as reported for the synthesis of **1**, and afforded crystalline (−)-2,3-methylenedioxy-2,3-didemethyoxycolchicine (**10**), identical in every respect with the natural alkaloid. The signal of the methylenedioxy protons showed as a singlet at δ 5.96 (Table I). This revealed that the methylenedioxy group in the natural alkaloid **10**, in contrast to compound **1**, had indeed an arrangement where both protons were undistinguishable by ^{1}H NMR.

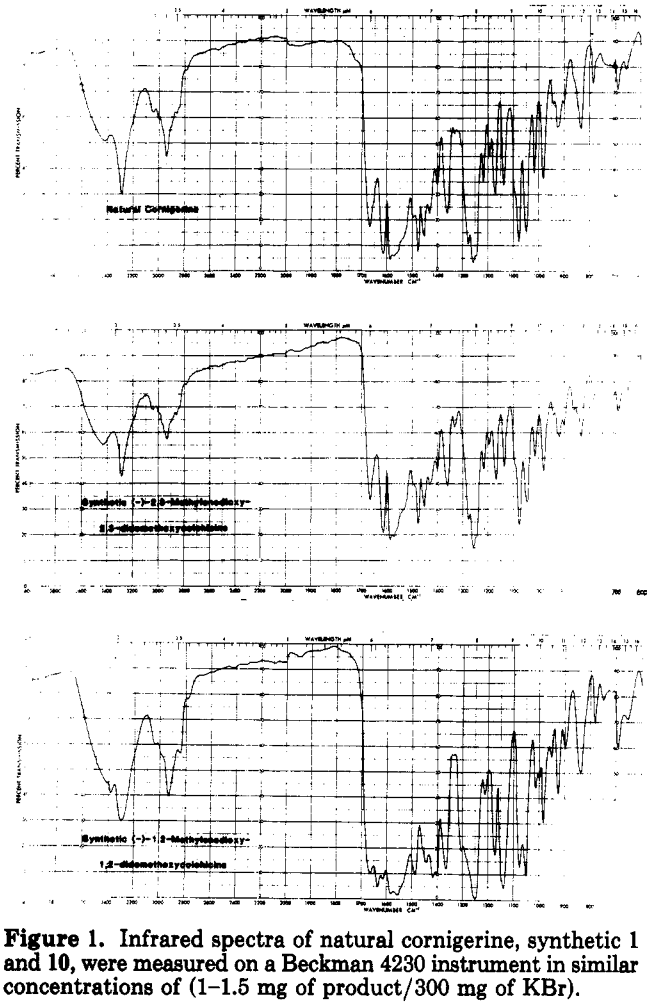

Figure 1. Infrared spectra of natural cornigerine, synthetic **1** and **10**, were measured on a Beckman 4230 instrument in similar concentrations of (1–1.5 mg of product/300 mg of KBr).

Cornigerine (**10**) prepared from colchicine showed identical optical properties, demonstrating that it has the same absolute configuration at C-7 as cholchicine (**2**). Synthetic (−)-2,3-methylenedioxy-2,3-didemethoxycolchicine (**10**) proved to be indeed identical in every respect (melting and mixture melting point, optical behavior, TLC analysis, ^{1}H NMR and IR in KBr; see Figure 1) with natural cornigerine. Synthetically prepared (−)-1,2-(methylenedioxy)-1,2-didemethoxycolchicine (**1**) differed significantly from the natural alkaloid in its optical behavior (Experimental Section), ^{1}H NMR spectrum (see Table I), and IR spectrum in KBr (see Figure 1, bottom). The originally proposed structure **1** for natural cornigerine therefore has to be changed to **10**.

Experimental Section

Melting points (corrected) were determined on a Thomas-Hoover capillary apparatus. Elemental analyses were performed by the Section on Microanalytical Services and Instrumentation of this Laboratory. IR spectra were obtained on a Beckman 4230 and mass spectra on a Hitachi Perkin-Elmer RMU-6E (70 eV) and a Finnigan 1015 (CI). ^{1}H NMR spectra were obtained (using tetramethylsilane at δ 0.0 as an internal reference) on a Varian HR-220, Bruker WP60, or JEOL FX-100 spectrometer. Optical rotations were measured on a Perkin-Elmer 241 MC polarimeter in chloroform solution. For TLC, SiO_2 GF or Al_2O_3 GF plates (Analtech) were used. Column chromatography was carried out by using basic Al_2O_3 (Alfa Division Ventron Corp.) or silica gel 60 (E. Merck). Detection was done by UV at 254 nm, by I_2 vapor, and by ethanolic $FeCl_3$ solution (1%).

1,2-Didemethylcolchicine (6).[14] A solution of colchicine (**2**; 2 g, 5 mmol) in 7.5 mL of concentrated H_2SO_4 was stirred at 55 °C for 3 h and then 2 h at 85–90 °C. The reaction time at 85–90 °C is rather critical to avoid the formation of further demethylated products. All reactions in concentrated H_2SO_4 were monitored by TLC. A small drop of the reaction mixture was diluted with MeOH, applied to a SiO_2 plate, and developed in CH_2Cl_2/MeOH (90:10). The reaction mixture was cooled with ice, carefully

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(13) Potěšilová, H.; Hruban, L.; Šantavý, F. *Collect. Czech. Chem. Commun.* **1975**, *41*, 3146.

(14) This catechol is only sparingly soluble in commonly used solvents. The crude residues obtained after the extraction and evaporation of solvents retain solvents, making it difficult to ascertain a material balance.

adjusted with 2 N NaOH to pH 3–5, and extracted with CH_2Cl_2 followed by CH_2Cl_2/MeOH (9:1). The combined fractions were washed (brine), dried (Na_2SO_4), and evaporated. TLC analysis of this extract revealed the presence of a small amount of phenol **4** besides more polar material (dark blue color with $FeCl_3$). Crystallization from CH_2Cl_2/MeOH afforded 210 mg of 1,2-didemethylcochicine (**6**) as yellow needles: mp 239 °C dec; mass spectrum (CI and EI), *m/e* 371; NMR (100 MHz, Me_2SO-d_6) δ 1.84 (s, 3, CH_3CO) 1.9–2.4 (m, 4, CH_2CH_2), 3.80 (s, 3, 3-OCH_3), 3.84 (s, 3, 10-OCH_3), 4.36 (m, 1, C_7 H), 6.40 (s, 1, C_4H), 6.96 (d, 1, C_{11} H, J = 11 Hz), 7.12 (s, 1, C_8 H), 7.20 (d, 1, C_{12}H, J = 11 Hz), 8.41 (d, 1, NH, J = 8 Hz), 8.56 (br s, 2, OH).

Analogous treatment of 1-demethylcolchicine (**3**)[7] resulted in a very similar reaction mixture. In this case, **4** was lacking, but a small amount of starting material **3** was present.

1,2-Diacetyl-1,2-didemethylcolchicine (7). A sample of 200 mg of slightly impure **6** was acetylated in 5 mL of pyridine with 1 mL of acetic anhydride at room temperature over 24 h. After the solution was adjusted to pH 1 with hydrochloric acid, the product was extracted with CH_2Cl_2. After the mixture was washed (brine) and dried (Na_2SO_4), evaporation of solvent gave a brown residue which crystallized from ethyl acetate. Recrystallization from ethyl acetate/ether gave 30 mg of **7** as yellowish crystals: mp 233–235 °C dec; mass spectrum (CI), *m/e* 455; NMR (100 MHz, $CDCl_3$), δ 1.95 (s, 3, *N*-$COCH_3$), 2.16 and 2.29 (2 s, 2 × 3, 1-$OCOCH_3$ and 2-$OCOCH_3$), 2.0–2.7 (m, partly hidden, CH_2CH_2), 3.86 (s, 3, 3-OCH_3), 3.96 (s, 3, 10-OCH_3), 4.64 (m, 1, C_7H), 6.70 (s, 1, C_4H), 6.74 (d, 1, C_{11} H, J = 11 Hz), 7.08 (d, 1, C_{12} H, J = 11 Hz), 7.49 (s/m, 2, C_8 H and NH).

1,2-(Methylenedioxy)-1,2-didemethoxycolchicine (1). A mixture of 170 mg of 1,2-diol **6** or 1,2-diacetate **7**, 500 mg of K_2CO_3, and 0.2 mL of bromochloromethane in 5 mL of 1-methyl-2-pyrrolidinone was stirred under N_2 for 2.5 h at 70 °C. After the mixture cooled, water was added and the solution extracted with CH_2Cl_2. The extracts were washed (brine), dried (Na_2SO_4), and evaporated. The oily residue containing some 1-methyl-2-pyrrolidinone was diluted with 150 mL of ether. The resulting precipitate was filtered off and crystallized from acetone to give 80 mg of slightly yellow crystals of **1**: mp 273–274 °C dec; mass spectrum (CI), *m/e* 383; IR ($CHCl_3$) 1672, 1640, 1616, 1593, 1560 cm^{-1}; NMR (100 MHz, $CDCl_3$) δ 1.93 (s, 3, CH_3CO), 2.0–2.6 (m, 4, CH_2CH_2), 3.92 (s, 3, 3-OCH_3), 3.96 (s, 3, 10-OCH_3), 4.7 (m, 1, C_7H), 5.91/6.05 (AB q, 2, OCH_2O, apparent d, J_{gem} < 2 Hz), 6.36 (s, 1, C_4H), 6.83 (d, 1, C_{11} H, J = 11 Hz), 7.33 (d, 1, C_{12} H, J = 11 Hz), 7.53 (s, 1, C_8H), 7.64 (br s, 1, NH). $[\alpha]^{25}_{D}$ −108° (*c* 1.002, $CHCl_3$). Anal. Calcd for $C_{21}H_{21}NO_6$: C, 65.79; H, 5.52; N, 3.65. Found: C, 65.58; H, 5.75; N, 3.63.

This material was different from natural cornigerine by IR (KBr) (see Figure 1), NMR ($CDCl_3$), and optical rotation. It could not, however, be separated from natural cornigerine by TLC.

2,3-Dimethylcolchicine (8).[14] A solution of 900 mg of 3-demethylcolchicine (**5**) in 3 mL of concentrated H_2SO_4 was heated at 85–90 °C with TLC monitoring (see preparation of **6**). The reaction time may vary from 1.5 to 2.5 h. Under cooling with ice the mixture was adjusted to pH 5 with 2 N NaOH and extracted once with 20 mL of CH_2Cl_2 to remove traces of starting material. The very polar **8** was then extracted six times with a total of 300 mL of CH_2Cl_2/2-propanol (7:3). After the mixture was dried (Na_2SO_4) and evaporated a brown glassy residue (220 mg) was obtained which could not be crystallized (in accord with the literature[13]). The structure of the crude compound was confirmed by the appearance of only two methoxy signals in the NMR (60 MHz, Me_2SO-d_6) at δ 3.42 (1-OCH_3) and 3.84 (10-OCH_3) [lit.[13] ($CDCl_3$) δ 3.63 and 4.01]; mass spectrum (EI), *m/e* (relative intensity) 371 (100), 343 (M^+ − CO, 13), 284 (M^+ − CO − H_2NCOCH_3, 93).

2,3-Diacetyl-2,3-didemethylcolchicine (9). A small amount of 2,3-diol **8** (40 mg) was characterized by acetylation by use of the conditions for the preparation of 1,2-diacetyl compound **7**. The material was obtained as a glass (TLC pure) which could not be crystallized: mass spectrum (CI), *m/e* 455; NMR (100 MHz, $CDCl_3$) δ 1.96 (s, 3, N − $COCH_3$), 2.31 and 2.33 (2 s, 2 × 3, 2-$OCOCH_3$, and 3-$OCOCH_3$), 3.57 (s, 3, 1-OCH_3), 3.99 (s, 3, 10-OCH_3).

2,3-(Methylenedioxy)-2,3-didemethoxycolchicine, Cornigerine (10). A stirred mixture of 250 mg of 2,3-diol **8**, 1 g of K_2CO_3, 1 mL of bromochloromethane, and 5 mL of 1-methyl-2-pyrrolidinone was heated under an N_2 atmosphere to 70 °C for 2.5 h. After the mixture was cooled, water was added and the solution extracted with CH_2Cl_2. The combined extracts were washed (brine), dried (Na_2SO_4), and evaporated. Residual 1-methyl-2-pyrrolidinone was stripped at high vaccum. The slight brown residue crystallized on addition of ether to give 215 mg of crude 2,3-methylenedioxy compound. The crude material was dissolved in CH_2Cl_2 and purified by filtration over basic Al_2O_3 (activity I, 1.5 cm i.d. × 12 cm column). After a 50 mL fraction of CH_2Cl_2 was discarded, the product was eluted with 50 mL of CH_2Cl_2/MeOH (99:1). Evaporation of solvents and crystallization from CH_2Cl_2/ether or ethyl acetate/ether gave pure 2,3-methylenedioxy compound **10**: mp 263–264 °C dec (natural cornigerine, mp 256 °C,[14] mmp 263–264 °C dec); mass spectrum (EI), (relative intensity) 383 (M^+, 41), 355 (M^+ − CO, 15), 340 (M^+ − $COCH_3$, 12), 296 (M^+ − CO − H_2NCOCH_3, base peak); IR, see Figure 1; NMR (60 MHz, $CDCl_3$) δ 1.96 (s, 3, CH_3CO), 1.8–2.8 (m, 4, CH_2CH_2), 3.77 (s, 3, 1-OCH_3), 3.97 (s, 3, 10-OCH_3), 4.6 (m, 1, C_7H), 5.96 (s, 2, OCH_2O), 6.41 (s, 1, C_4H), 6.80 (d, 1, C_{11} H, J = 11 Hz), 7.20 (d, 1, C_{12} H, J = 11 Hz), 7.48 (s, 1, C_8H), 7.58 (d, 1, NH, J = 6 Hz); $[\alpha]^{22}_{D}$ −149.8° (*c* 0.623, $CHCl_3$) (lit.[2] $[\alpha]^{20}_{D}$ −150 ± 2° (*c* 0.631, $CHCl_3$)). Anal. Calcd for $C_{21}H_{21}NO_6 \cdot 0.5H_2O$: C, 64.28; H, 5.65; N, 3.57. Found: C, 64.26; H, 5.83; N, 3.34.

This material was identical by melting and mixture melting point, IR (KBr), NMR ($CDCl_3$), and optical rotation ($CHCl_3$) with a reference sample of natural cornigerine.[15]

Registry No. **1**, 78249-43-1; **2**, 64-86-8; **3**, 3464-68-4; **4**, 7336-36-9; **5**, 7336-33-6; **6**, 78231-83-1; **7**, 78231-84-2; **8**, 57866-21-4; **9**, 78231-85-3; **10**, 6877-25-4.

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(15) We thank Professor Šantavý, Institute of Medical Chemistry, Medical Faculty of the Palacky University, Olomouc, Czechoslovakia, for having provided us with a sample of natural cornigerine. This sample in our hands showed a corrected melting point of mp of 262–263 °C.