diation with a graphite monochromator (4104 reflections, 1533 observed). Only the latter data set was used in the refinement reported here.

Acknowledgments. This research was supported in part by a USPHS Grant (CA-10906) to the Medical Foundation of Buffalo.

Registry No.—**1**, 570-74-1; **2**, 56114-17-1; hydrogen chloride, 7647-01-0.

Supplementary Material Available. Tables of coordinates, thermal parameters, structure factors, and packing diagrams (Figures 5 and 6) will appear following these pages in the microfilm edition of the volume of the journal. Photocopies of the supplementary material from this paper only or microfiche (105 × 148 mm, 24× reduction, negatives) containing all of the supplementary material for the papers in this issue may be obtained from the Business Office, Books and Journals Division, American Chemical Society, 1155 16th Street, N.W., Washington, D.C. 20036. Remit check or money order for $4.50 for photocopy or $2.50 for microfiche, referring to code number JOC-75-2956.

References and Notes

(1) (a) Medical Foundation of Buffalo; (b) University of Virginia.
(2) J. Mauthner, *Monatsh. Chem.*, **27**, 305 (1906); *Chem. Abstr.*, **2**, 659 (1908); *Monatsh. Chem.*, **30**, 635 (1909); *Chem. Abstr.*, **4**, 1158 (1910).
(3) (a) L. F. Fieser and M. Fieser, "Steroids", Reinhold, New York, N.Y., 1959, p 33; (b) C. W. Shoppee, "Chemistry of the Steroids", Butterworths, London, 1958, p 17.
(4) D. H. R. Barton and C. H. Robinson, *J. Chem. Soc.*, 3045 (1954).
(5) J. D. Bernal, D. Crowfoot, and I. Fankuchen, *Philos. Trans. R. Soc. London, Ser. A*, **239**, 135 (1940); D. Crowfoot in "Vitamins and Hormones", Vol. II, Academic Press, New York, N.Y., 1944, p 409.
(6) T. Kon, *J. Pharm. Soc. Jpn.*, **64**, 15 (1944); *Chem. Abstr.*, **45**, 5703 (1951).
(7) (a) R. C. Fort, Jr., R. E. Hornish, and G. A. Liang, *J. Am. Chem. Soc.*, **92**, 7558 (1970); (b) R. C. Fort, Jr., in "Carbonium Ions", Vol. IV, G. A. Olah and P. v. R. Schleyer, Ed., Wiley-Interscience, New York, N.Y., 1973, Chapter 29; (c) G. E. Gream, *Aust. J. Chem.*, **25**, 1051 (1972); (d) K. B. Becker, A. F. Boschung, M. Geisel, and C. A. Grob, *Helv. Chim. Acta*, **56**, 2743 (1973).
(8) (a) R. M. Carlson and R. K. Hill, *J. Org. Chem.*, **34**, 4178 (1969); (b) F. A. Carey and H. S. Tremper, *ibid.*, **36**, 758 (1971); F. D. Greene and N. N. Lowry, *ibid.*, **32**, 875 (1967).
(9) J. A. Marshall, *Acc. Chem. Res.*, **2**, 33 (1969).
(10) (a) R. F. Zurcher, *Helv. Chim. Acta*, **44**, 1380, 1755 (1961); **46**, 2054 (1963); (b) J.-C. Jacquesy, J.-M. Lehn, and J. Levisalles, *Bull. Soc. Chim. Fr.*, 2444 (1961); (c) J.-C. Jacquesy, R. Jacquesy, J. Levisalles, J.-P. Pete, and H. Rudler, *ibid.*, 2224 (1964); (d) E. R. Malinowski, M. S. Manhas, G. H. Muller, and A. K. Bose, *Tetrahedron Lett.*, 1161 (1963).
(11) Unpublished work of M. Wurth cited in ref 9. We thank Professor Marshall for providing copies of these NMR spectra.

Isolation and Structure of 1-Hydroxy-7-methoxy-4-isopropyl-1,6-dimethyl-2(1*H*)-naphthalenone from Cotton

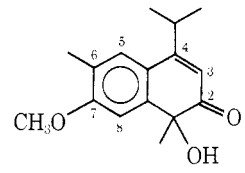
Peter W. Jeffs* and David G. Lynn

P. M. Gross Chemical Laboratory, Duke University, Durham, North Carolina 27706

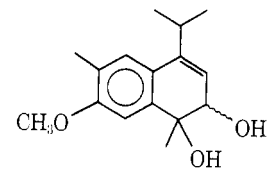
Received May 6, 1975

Previous work from this laboratory has been concerned with attempts to identify the components in the cotton plant *Gossypium hirsutum* which are implicated in producing the clinical symptoms of the disease byssinosis.[1] In this earlier study we reported preliminary characterization of a column chromatographic fraction from cotton dust or cotton bracts which exhibited chemotactic activity with human polymorphonucleur cells. Purification of this fraction by preparative layer chromatography afforded 70 μg of a compound exhibiting a strong yellow-green fluorescence under 254-nm ultraviolet light which was tentatively assigned a molecular weight of 260 from mass spectral examination.

Reisolation of 20 mg of this compound has now permitted its characterization as 1-hydroxy-7-methoxy-4-isopropyl-1,6-dimethyl-2(1*H*)-naphthalenone (**1**).



1



2

The initial isolation studies of the fluorescent component were directed toward examination of the volatile fraction on the basis that steam treatment of cotton is reported to reduce or destroy the byssinosis factor when measured by the response of susceptible workers.[2] The yields of the fluorescent fraction obtained by this procedure were extremely low (~10 μg/kg) and alternative sources and methods of isolation were examined. Using the fluorescence properties as a guide to the presence of active material, it was found that aged cotton bracts contained more than fresh bracts and that fresh leaves and stems contained only insignificant amounts of this material.

Extensive purification of an aqueous acetone extract of aged cotton bracts by column chromatography afforded a crude fraction containing the yellow-green fluorescent component. Preparative layer chromatography of this fraction on silica gel in chloroform–5% methanol gave a pure compound, mp 100–102°, which proved identical in its chromatographic and mass spectral characteristics with that previously obtained.

An exact mass measurement of the molecular ion, *m/e* 260, from the electron impact (EI) spectrum established the molecular formula as $C_{16}H_{20}O_3$. Verification that the ion *m/e* 260 was indeed the molecular ion was obtained from the chemical ionization (CI) spectrum, which showed a quasimolecular ion at *m/e* 261 (100).

The CI spectrum was remarkably simple in that in addition to the QM^+ ion the only ions of significant intensity appear at $(QM^+ + 1)$, $(QM^+ - OH)$, and $(QM^+ - H_2O)$ and account for 90% of the total ion current. In contrast, the high-resolution EI spectrum contained many ions of which those corresponding to $M^+ - CH_3$ at *m/e* 245 and $M^+ - CO$ at *m/e* 232 were present. The loss of CH_3 from the *m/e* 232 ion to a fragment ion of *m/e* 217, when taken in conjunction with the appearance of an ion at *m/e* 202, suggested the presence of two methyl groups and a ketone. In addition, the base peak at *m/e* 189 ($C_{12}H_{13}O_2$) and an ion at *m/e* 175 ($C_{11}H_{11}O_2$) were the only other ions of any significance at high mass. The occurrence of absorption bands at 1670 and 3490 cm^{-1} in the ir spectrum of **1** confirmed the presence of an unsaturated carbonyl and hydroxyl functions, respectively.[3] The invariance of the OH band at 3490 cm^{-1} with increasing dilution in CCl_4 solution demonstrated that the OH group was intramolecularly hydrogen bonded and the frequency of the absorption was consistent with that expected for an α-hydroxy ketone.[4]

In agreement with the fluorescence properties of **1**, the uv spectrum in cyclohexane showed evidence for extended conjugation with bands at 222 nm (ε 9100), 227 (10,400), 252 (10,800), 258 (11,300), 335 (4730), and 365 (2730). An indication that the latter was consistent with a β-aryl-α,β-unsaturated ketone chromophore was supported by a comparison of its spectrum in ethanol, which was much less detailed than in cyclohexane, with the uv of other 2(1*H*)-naphthalenones[5] and by the marked change in the uv spec-

trum associated with the reduction of 1 to its dihydro derivative 2 with sodium borohydride (see Experimental Section).

The 100-MHz ^1H and the ^{13}C spectra of the fluorescent component provided considerable structural information. These data when considered in conjunction with other spectral results were sufficient to permit an assignment of structure. A pair of 3 H doublets (J = 7.0 Hz) at δ 1.24 and 1.26 which were coupled (DNMR) to a 1 H multiplet at δ 3.14 was observed in the ^1H NMR spectrum of 1 and indicated the presence of an isopropyl group in which the methyl groups are diastereotopic. Carbon resonances[6] at δ 21.8 and 22.1 for the methyls and δ 28.2 for the methine carbon supported the presence of an isopropyl group.[8] Methyl resonances in the proton spectrum occurred as singlets at δ 1.52, 2.22, and 3.88 and, in conjunction with the corresponding carbon resonances at 33.4,[8] 16.1,[9] and 55.5[9] ppm, allowed their assignment to a methyl on a carbon containing an hydroxyl group, an aromatic methyl, and an aromatic methoxyl. One-proton singlets at δ 6.08, 7.23, and 7.34 were attributable to the α hydrogen of an α,β-unsaturated ketone and aromatic hydrogens in a para relationship, respectively. A DNMR experiment showed that the low-field aromatic signal was slightly broadened through coupling to the methyl at δ 2.22. Support for the presence of a β-substituted enenone system was provided by the carbon signals at 125 and 164.1 ppm. The highly deshielded position of the latter is in agreement with the expected position of the β carbon of a β-substituted α,β-unsaturated ketone system.[10]

Although the foregoing spectral data are consistent with structure 1, there are several alternatives which cannot be excluded from consideration. These structures differ from 1 by interchanging the position of the aromatic substituents and the isopropyl group. Two of these alternative structures could be discounted from the observation that compound 1 is recovered from an acidic solution and therefore it is not in accord with the properties expected for a β-diketone enol ether.[11] The structural possibilities are reduced to that proposed and the biogenetically less attractive alternative in which the position of the methoxyl and aromatic methyl are interchanged.

A decision in favor of structure 1 was provided by a lanthanide shift study. The small coupling observed between the methyl group on the aromatic ring and the aromatic hydrogen associated with the signal of δ 7.34 indicates that the aromatic signal at δ 7.23 originates from the hydrogen ortho to the methoxyl group. During additions of incremental amounts of $Eu(fod)_3$ to a sample of 1, the largest $\Delta\delta$ was observed for the methyl signal of δ 1.52 in agreement with the expected complexation of the europium at the hydroxyl group.[12] Of the two aromatic hydrogen signals, the low-field signal at δ 7.34 remains essentially unchanged upon addition of $Eu(fod)_3$ while the signal at δ 7.23 is shifted appreciably. This finding clearly is consistent with structure 1 and suffices to rule out the alternative structure in which the positions of the methyl and methoxyl on the aromatic ring are reversed. This compound represents a further example of the sesquiterpenes found in cotton which are based upon the cadanane skeleton.[13] Despite the fact that structure 1 is chiral, examination of the CD and ORD spectra indicated that 1 is produced in the racemic form. At this stage the role of 1 in causing the onset of symptoms associated with byssinosis is under active investigation. The increased incidence of 1 in aged bracts and its absence from fresh plant material may indicate that its occurrence is linked to the presence of contaminating microorganisms. A recent report[14] has described the occurrence of isohemigossypol as a phytoalexin in cotton plants infected with *Verticillium dahliae*.

Experimental Section

Melting points were determined on a Kofler hot-stage apparatus. Infrared spectra were obtained in chloroform solutions on a Perkin-Elmer Model 621 spectrophotometer. Ultraviolet spectra were measured in cyclohexane unless otherwise noted on a Cary 15 spectrophotometer. Proton nuclear magnetic resonance (NMR) spectra were recorded at 90 MHz on a Bruker HFX-90 and at 100 MHz on a Jeol MH-100 in $CDCl_3$ solution containing Me_4Si. The Fourier transform carbon-13 spectrum was obtained on a Jeol PS-100 through the courtesy of I. Carrol, Research Triangle Institute. The spectrometer was operated at 25.0292 MHz and was equipped with a high-sensitivity insert. The Fourier transforms were based upon 8K data points and employed the absorption spectrum. A sample concentration of 18 mg in 0.3 ml of $CDCl_3$ (containing 1% Me_4Si) contained in a 5-mm sample tube was used. The spectrum was recorded with noise decoupling in the proton region. A total of 64K transients were accumulated under conditions using a pulse width of 12.5 μsec and a repetition rate of 1.0 sec. High-resolution mass spectra were obtained on an MS-902 at 70 eV using a direct insertion probe. Chemical ionization spectra were obtained on a Finnigan 3300 quadrupole mass spectrometer operating in the CI mode with electron energy of 125 eV at a source pressure of 500 μ of isobutane and a source temperature of 125°. Circular dichroism spectra were obtained on a Jasco ORD/CD spectropolarimeter. Gas–liquid chromatography (GLC) was carried out on a Hewlett-Packard 402 instrument with 8 ft × 0.125 in. glass columns packed with 3% OV-17, 3% QF-1, or 3% SE-30 on Gas-Chrom Q (100–120 mesh) operating at 200°.

Isolation. The cotton stems and bracts (3.65 kg) obtained from the inclined cleaner of a cotton mill were ground in a blender at high speed for ca. 1–2 min with acetone–water (85:15) (12 ml/g) and was then filtered over Celite. The filtrate was treated with lead acetate solution;[7] one part of the lead acetate solution was added to four parts of deionized water which was then added to an equal volume of the cotton filtrate, and the resulting suspension was filtered over Celite. This filtrate was extracted with an equal volume of chloroform, the organic layer was taken to dryness under reduced pressure, and the resulting oil (26 g) was applied to a column (27 × 2.75 in.) containing 1.3 kg of neutral alumina (activity 1). The column was eluted sequentially with benzene (3 l.), a continuous gradient of benzene–chloroform (6 l.), chloroform (3 l.), a continuous gradient of chloroform–methanol (6 l.), and methanol (3 l.). The desired yellow fluorescent material along with a blue fluorescent compound and second yellow fluorescent component were eluted with methanol. Purification of this fraction by preparative layer chromatography (PLC) on silica gel in $CHCl_3$–MeOH (20:1) gave a major component at R_f 0.93 which was further purified by PLC on silica gel in benzene–ether (4:6) to give 20 mg of 1 which crystallized slowly from CCl_4: mp 100–102°; ir ($CHCl_3$) 3490 (OH), and 1670 cm^{-1} (CO), the 3490-cm^{-1} band was not affected by dilution of the sample to concentrations of 1×10^{-4} M; uv (hexane) 222 nm (ϵ 9100), 227 (10,400), 252 (11,800), 258 (11,360), 335 (4730), 365 (2730); uv (95% EtOH) 228 (10,200), 248 (10,300), 339 (4300), 370 (sh); ^1H NMR (100 MHz) δ 1.24 (d, J = 7.0 Hz, 3), 1.26 (d, J = 7.0 Hz, 3), 1.52 (s, 3), 2.22 (s, 3), 3.14 (m, 1), 3.88 (s, 3), 6.08 (s, 1), 7.23 (s, 1), 7.34 (s, 1); ^{13}C NMR (25.029 MHz) 16.1, 21.8, 22.1, 28.2, 29.6, 33.9, 55.5, 75.6, 107.2, 114.7, 125.2, 125.5, 127.5, 145.4, 164.1 ppm (C=O signal was saturated under the operating conditions used); mass spectrum (70 eV) m/e (rel intensity) 260 (40, M^+), 245 (6, M − CH_3), 232 (30, M − 60), 217 (66, M − CO + CH_3), 202 (21, M − CO + $2CH_3$), 189 (100, $C_{12}H_{13}O_2$). GLC on QF-1 or SE-30 columns gave a single peak. Compound 1 was also isolated from cotton dust obtained from the floor sweepings of a cotton mill. A similar extraction of fresh leaves and bracts gave no evidence for the presence of 1.

Anal. Calcd for $C_{16}H_{20}O_3$: m/e 260.1412. Found: m/e 260.1416.

Borohydride Reduction of 1. A solution of 1 (2.89 mg, 11 μmol) in 2-propanol (1 ml) was added to a stirred solution of $NaBH_4$ (2 mg) in 2-propanol (1 ml) over a period of 45 min at room temperature. Preparative layer chromatography on silica gel in $CHCl_3$–MeOH (20:1) gave the dihydro compound 2: mp 172–174°; R_f 0.45; ir 3558 cm^{-1} (OH), no C=O; uv 258 nm (3200), 284 (3870); mass spectrum m/e (rel intensity) 262 (44, M^+), 244 (23, M − 18), 219 (47, M − 43), 201 (53), 159 (100).

Anal. Calcd for $C_{16}H_{22}O_3$: m/e 262.1568. Found: m/e 262.1564.

Attempted Hydrolysis of 1. A solution of 1 (3 mg) in dioxane (2.5 ml) was heated with 2.5 ml of 1 *N* HCl and the reaction was monitored by GLC on a SE-30 column periodically for 1 hr. No change in the intensity of the peak associated with 1 was observed during this time period.

Acknowledgments. This research was supported through the award of a training grant in Environmental Sciences from the National Institute of Environmental Health Sciences (5-TOJ-ES-00124) and a Biological Sciences Support Grant to Duke University. High-resolution mass spectral results were provided by the Research Triangle Institute of Mass Spectrometry through the courtesy of Dr. David Rosenthal and Mr. Fred Williams. Dr. P. E. Sasser and Mr. W. Taylor, Cotton Inc., kindly provided the cotton plant material and samples of cotton dust.

Registry No.—1, 56051-00-4; 2, 56051-01-5.

References and Notes

(1) W. S. Lynn, Munoz, J. A. Campbell, and P. W. Jeffs, *Ann. N.Y. Acad. Sci.*, **221**, 163 (1974).
(2) G. Taylor, A. A. E. Massoud, and F. Lucas, *Br. J. Ind. Med.*, **28**, 143 (1971).
(3) The bands at 1740 and 1700 cm^{-1} reported earlier on the 70-μg sample were absent in the purified material.
(4) M. Tichy, *Adv. Org. Chem.*, **5**, 115 (1965).
(5) R. M. Dodson, J. R. Lewis, W. P. Webb, and E. Wenkert, *J. Am. Chem. Soc.*, **83**, 938 (1961); E. Wenkert, R. D. Youssefyeh, and R. G. Lewis, *ibid.*, **82**, 4675 (1960).
(6) The small quantity of compound available precluded the possibility of obtaining ^{13}C off-resonance or partially reluxed spectra. The assignments are based upon chemical shift theory[7] and/or comparison with suitable models.
(7) J. W. Emsley, J. Feeney, and L. H. Sutcliffe, "High Resolution Nuclear Magnetic Resonance Spectroscopy", Vol. 2, Pergamon Press, Elmsford, N.Y., 1966; E. Lippmaa, T. Pekh, K. Anderson, and C. Rappe, *Org. Magn. Reson.*, **2**, 109 (1970).
(8) L. F. Johnson and W. C. Jankowski, ^{13}C NMR Spectra", Wiley-Interscience, New York, N.Y., 1972, Spectra No. 314, 352, 354, and 436.
(9) J. B. Stothers, "Carbon-13 NMR Spectroscopy", Academic Press, New York, N.Y., 1972; G. C. Levy and G. L. Nelson, "Carbon-13 Nuclear Magnetic Resonance for Organic Chemists", Wiley-Interscience, New York, N.Y., 1972.
(10) E. Wenkert, B. Chauncy, K. G. Dave, A. R. Jeffcoat, F. M. Schell, and H. P. Schenk, *J. Am. Chem. Soc.*, **95**, 8427 (1973).
(11) V. Arkley, J. Attenburrow, G. I. Gregory, and T. Walker, *J. Chem. Soc.*, 1260 (1962).
(12) J. K. M. Sanders and D. H. Williams, *Chem. Commun.*, 422 (1970).
(13) Dr. P. J. Wakelyn, National Cotton Council of America, has informed us that he has isolated compound 1 from cotton and has independently arrived at its structure. Note Added in Proof. D. Seipanovic, P. J. Wakelyn, and A. A. Bell, *Phytochem.*, **14**, 1041 (1975).
(14) A. S. Sandykov, L. V. Melitskii, A. K. Karimdzhanov, A. I. Ismailov, R. A. Mukhamedova, M. Kh. Avazkhodzhaev, and F. G. Kamaev., *Dokl. Akad. Nauk SSR*, **218**, 1472 (1974); *Chem. Abstr.*, **82**, 82996 (1975).

α-Chlorination of Aliphatic Acids by Molecular Chlorine

Yoshiro Ogata,* Taira Harada, Kazuo Matsuyama, and Toshinori Ikejiri

Contribution No. 214 from Department of Applied Chemistry, Faculty of Engineering, Nagoya University, Chikusa-ku, Nagoya, Japan

Received March 21, 1975

We have previously reported that enolizing catalysts such as H_2SO_4, HCl, or $FeCl_3$ together with *m*-dinitrobenzene as a radical trapper increase the ratio of α- vs. β-chlorination of propionic acid by molecular chlorine.[1]

As an extension of this study concerning the effect of catalyst, the authors discovered that aliphatic acids can be effectively α-chlorinated in the presence of molecular oxygen as a radical trapper, which was pointed out in the Cl_2 addition to double bond[2] and in the phosphorus chloride catalyzed chlorination of alkanes.[3] The significant effect of molecular oxygen on the α-chlorination in the presence of 95% H_2SO_4 is listed in Table I.

Table I
Effect of Radical Trappers on the Chlorination Product of Butyric Acid[a]

Radical trapper	(mol %)	Registry no.	Yield, % α-Chloro acid	Yield, % β-Chloro acid
m-$C_6H_4(NO_2)_2$	(7.0)	99-65-0	6.7	1.4
O_2[b]		7782-44-7	22.3	0

[a] Chlorine gas was continuously introduced into the substrate at the flow rate of 200 ml/min at 120° for 3 hr in the dark with initial amounts of butyric acid (0.2 mol) and H_2SO_4 (0.02 mol). [b] The flow rate of O_2 was 200 ml/min.

The authors discovered also that the yield of α-chlorinated product further increases by the use of chlorosulfonic acid instead of concentrated H_2SO_4 as an enolizing catalyst. The remarkable effect of chlorosulfonic acid is probably due to the ability of formation of a more homogenous mixture and to the stronger acidity compared with that of H_2SO_4. Moreover, the authors examined the ability of some radical trappers and found that chloranil has an apparent effect on the α-chlorination.[4] These results are summarized in Table II for the chlorination of isovaleric acid as a substrate.

Table II
Effect of Chlorosulfonic Acid and Chloranil on Chlorination of Isovaleric Acid[a]

O_2:Cl_2 mol ratio[b]	Chloranil, mmol	Enolizing catalyst (mmol)	Yield, % α-Chloro acid	Yield, % β-Chloro acid
1:2	0.04	95% H_2SO_4 (171)	23.0	1.2
1:2	0.04	$ClSO_3H$ (60)	70.6	0.0
1:2	0	$ClSO_3H$ (60)	72.8	0.0
No O_2	0	$ClSO_3H$ (60)	41.6	2.7
No O_2	3	$ClSO_3H$ (60)	65.6	7.3

[a] Dark reaction at 140° for 3 hr with an initial amount of isovaleric acid of 600 mmol. [b] The flow rates of Cl_2 and O_2 gas were 100 and 50 ml/min, respectively.

Table II shows that the most effective α-chlorination of isovaleric acid by molecular chlorine in the dark is possible in the presence of chlorosulfonic acid, molecular oxygen, and chloranil at 140°. The chlorination in the absence of these three addenda gave a mixture of several chloro acids.

Various aliphatic acids can be similarly α-chlorinated in the presence of the above catalysts ($ClSO_3H$, O_2, and chloranil) to give its corresponding α-chloro acids alone in excellent yields (Table III). No β-chloro acid is detected except with isobutyric, *n*-butyric, and isocaproic acid, which produce only a trace of the corresponding β-chloro acid. It is of interest to note that the α-chlorination is predominant even in the presence of tertiary hydrogen, e.g., isovaleric acid. These products were characterized on the basis of boiling point, ir, and NMR spectra of the corresponding methyl ester which is prepared by H_2SO_4-catalyzed esterification with a mixture of methyl alcohol and ethylene dichloride[5] (Table IV).

The authors already proposed that the acid-catalyzed chlorination of aliphatic acid by molecular chlorine in the dark may proceed via the ionic chlorination of enolized aliphatic acid, $RCH{=}C(OH)_2$, where the radical trappers minimize the radical chlorination.[1,6] The above results