violet spectra. Mrs. Iris J. Siewers and Miss Alice M. Bernardi, National Heart Institute, kindly determined the infrared spectra.
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[CONTRIBUTION FROM THE LABORATORY OF CHEMICAL PHARMACOLOGY, NATIONAL CANCER INSTITUTE, NATIONAL INSTITUTES OF HEALTH, PUBLIC HEALTH SERVICE, FEDERAL SECURITY AGENCY]

Components of Podophyllin. IX. The Structure of the Apopicropodophyllins[1,2]

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The constitution of α-apopicropodophyllin has been established as in formula I. Structure IIa for β-apopicropodophyllin is favored over IIb on spectroscopic evidence. A new isomer, γ-apopicropodophyllin (III), has been obtained and proven to be structurally identical with a known synthetic compound.

The revival of interest in the chemistry of podophyllotoxin and picropodophyllin[3,4] has motivated a renewed study of some of their derivatives, and especially of the apopicropodophyllins, which had been investigated in the past, but about whose precise structure little was known. Some new evidence which has now been obtained forms the subject of the present communication.

Borsche and Niemann[5] observed that when picropodophyllin,[4] $C_{22}H_{22}O_8$, was treated with acetic anhydride containing a trace of sulfuric acid, it underwent dehydration to a compound, $C_{22}H_{20}H_7$, which they named apopicropodophyllin. They were unable to purify the crude reaction product, which melted at varying temperatures ranging up to 237°, by recrystallization from organic solvents, but obtained material melting consistently at 214–216° by boiling it with acetic anhydride containing some sodium acetate. Späth, Wessely and Kornfeld[6] prepared what appeared to be the same product, m.p. 216°, by prolonged boiling of picropodophyllin with acetic anhydride; their material, however, had $[\alpha]^{22}D$ +75.31°, while Borsche's was reported to be optically inactive. It was found subsequently by Robertson and Waters[7] that the purified substance isolated by the previous investigators was not identical with the primary dehydration product. The crude material isolated in the reaction of picropodophyllin with acetic anhydride and sulfuric acid usually afforded needles, m.p. 236–237°, when crystallized repeatedly from ethanol–acetic acid, then from ethyl acetate, but in some instances further recrystallization gave a product which melted at 244–245° and had $[\alpha]^{21}_{5461}$ −17.5°. When this compound, which Robertson and Waters named α-apopicropodophyllin, was heated in acetic anhydride with pyridine or sodium acetate, it was converted to an isomer, named β-apopicropodophyllin, m.p. 216°, $[\alpha]^{21}_{5461}$ +117.6°. Saponification of either isomer yielded the same hydroxy acid, which melted at 174° with loss of water, forming again α-apopicropodophyllin. Drake and Price[3] also prepared β-apopicropodophyllin, m.p. 214.0–215.4°, by the pyrolysis of picropodophyllin benzoate.

The difficulty of obtaining pure α-apopicropodophyllin indicates that it is rather unstable and quite easily converted to the β-isomer. This conversion has previously been accomplished by the use of weakly basic reagents, such as the ones employed by Robertson and Waters,[7] and has been carried out in this Laboratory by the use of piperidine in glacial acetic acid, thus affording a practically quantitative yield of pure β-apopicropodophyllin m.p. 219–220° (in some cases 220–221° after recrystallization); $[\alpha]^{20}D$ +102° (chloroform). It appears, however, that the presence of only traces of alkaline material also produces at least partial isomerization. Thus, when α-apopicropodophyllin was dissolved in boiling ethanol, and the ultraviolet absorption spectrum of the solution studied after cooling, it was found that partial conversion to the β-isomer had taken place. Reproducible results, indicating that the spectrum was actually that of pure α-apopicropodophyllin, could be obtained by dissolving the compound in chloroform and diluting the solution with ethanol, or, preferably, with ethanol containing a small amount of hydrochloric acid. The observation that the isomerization of the α- to the β-isomer did not occur in the presence of mineral acid permitted for the first time obtaining consistently pure α-apopicropodophyllin, m.p. 243–245°, $[\alpha]^{20}D$ −18° (chloroform), by recrystallizing the crude dehydration product from glacial acetic acid containing a trace of hydrochloric acid. The conversion of α- to β-apopicropodophyllin also took place when the former was heated for a short time above its melting point.

The hydroxy acid, m.p. 174°, which Robertson and Waters[7] had obtained from both the α- and β-apolactones, appeared to be related to the α-isomer since the latter was formed by relactonization. This was confirmed by its color reaction with sulfuric acid, which was identical with that given by α-apopicropodophyllin,[7] and also by the fact that chloroform solutions of both the acid and the α-apolactone rapidly decolorized bromine, while those of the β-apolactone did not. The relationship is substantiated by the close resemblance of the ultraviolet absorption spectrum (Fig. 1) of the acid (obtained from either α- or β-apopicro-

(1) Paper VIII: A. W. Schrecker and J. L. Hartwell, THIS JOURNAL, **74**, 5672 (1952).

(2) Presented in part before the Division of Medicinal Chemistry of the American Chemical Society, Chicago, Ill., September 6, 1950; Abstr. Papers Am. Chem. Soc., **118**, 18 M (1950).

(3) N. L. Drake and E. H. Price, THIS JOURNAL, **73**, 201 (1951).

(4) J. L. Hartwell and A. W. Schrecker, *ibid.*, **73**, 2909 (1951).

(5) W. Borsche and J. Niemann, *Ann.*, **494**, 126 (1932).

(6) E. Späth, F. Wessely and L. Kornfeld, *Ber.*, **65**, 1536 (1932).

(7) A. Robertson and R. B. Waters, *J. Chem. Soc.*, 83 (1933).

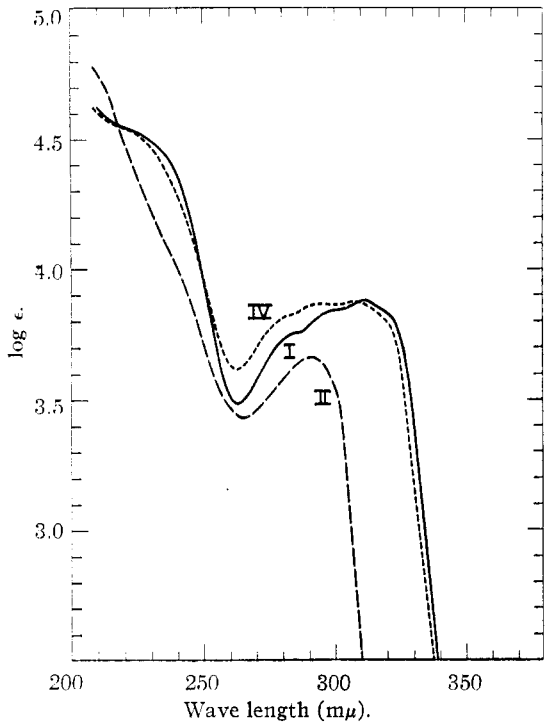


Fig. 1.—Ultraviolet absorption spectra of: ———, α-apopicropodophyllin (I); — — — —, β-apopicropodophyllin (II); -----, α-apopodophyllic acid (IV); solvent: 0.001 *N* HCl in 95% ethanol for I, 95% ethanol for II and IV.

podophyllin) with that of the α-apolactone, as contrasted with that of the β-apolactone. Thus the acid should properly be named α-apopodophyllic acid. That the ion of this acid is formed directly in the saponification of the lactones, and that therefore the acid is not produced upon acidification of a hypothetical β-apopodophyllate anion is demonstrated by the essential identity of the ultraviolet absorption spectrum of the solution resulting from the alkaline hydrolysis of β-apopicropodophyllin with that of the free acid. It is noteworthy that, while α-apopicropodophyllin is isomerized to β-apopicropodophyllin by basic reagents under conditions where the lactone ring structure is preserved, opening of the lactone ring results in formation of the acid corresponding to the α-isomer. The methyl ester of α-apopodophyllic acid, m.p. 172–173°, was prepared from the acid with diazomethane; its ultraviolet absorption spectrum was almost identical with that of the free acid. Reagents, such as sodium acetate, which isomerize the α-apolactone, do not affect the α-apo acid, while they convert its methyl ester to β-apopicropodophyllin.

When a solution of potassium α-apopodophyllate, prepared by dissolving α-apopicropodophyllin in 20% aqueous potassium hydroxide, was boiled for a day, partial isomerization took place, and a new hydroxy acid, named γ-apopodophyllic acid, could be isolated from the reaction mixture in low yield, after converting the unchanged α-apopodophyllic acid to an alkali-insoluble compound by treatment with iodine in bicarbonate solution as described below. This isomerization by means of strong alkali is analogous to the formation of the stable 3,4-dihydro-2-naphthoic acid from its less stable isomers, described by Derick and Kamm.[8] γ-Apopodophyllic acid, after purification, melted with effervescence when immersed at 200°, resolidified, and melted again at 249–250°. The corresponding lactone, named γ-apopicropodophyllin, was obtained by heating in the presence of mineral acid or by vacuum sublimation; after purification by chromatography on alumina it had m.p. 252–254°. Its chloroform solution did not decolorize bromine rapidly. Its ultraviolet (Figs. 2, 3) and infrared (Fig. 4) absorption spectra were

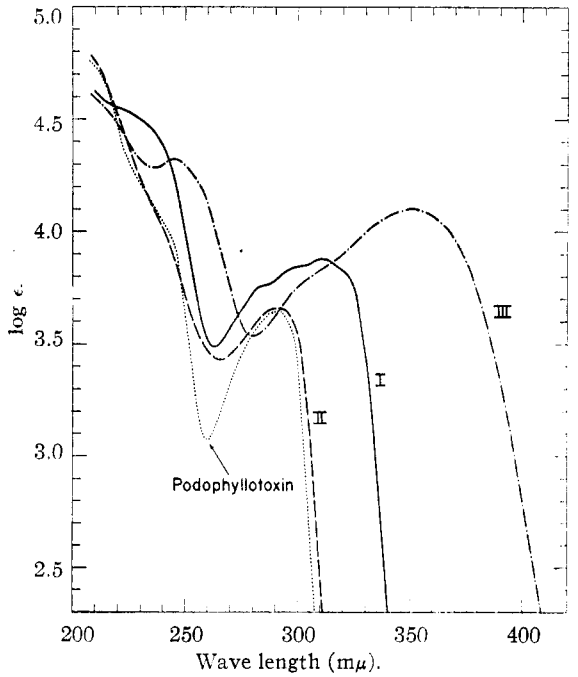


Fig. 2.—Ultraviolet absorption spectra of: ....., podophyllotoxin; ———, α-apopicropodophyllin (I); — — —, β-apopicropodophyllin (II); —·—·—, γ-apopicropodophyllin (III); solvent: 0.001 *N* HCl in 95% ethanol for I, 95% ethanol for the others.

identical in all respects with those of 6,7-methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-hydroxymethyl-3,4-dihydro-2-naphthoic acid lactone (III), which had been synthesized by Haworth and Richardson.[9,1] The ultraviolet spectra of γ-apopodophyllic acid and of the synthetic hydroxy acid[1] derived from III (Fig. 3) were also indistinguishable.

Some of the samples of γ-apopicropodophyllin obtained in repeated runs were found to be dextrorotatory, with a specific rotation of $[\alpha]_D$ +25 to +28° (observed rotations of around 1°), while others were devoid of optical activity. In one experiment, only optically active material was isolated. In other runs, fractionation of the crude γ-apopodophyllic acid gave first crops, which were converted to inactive lactone, and material obtained from the mother liquors, which was lactonized to dextrorotatory samples. The ultraviolet spectra of the latter were entirely identical with those of the inactive material; this eliminated the possibility

(8) C. G. Derick and O. Kamm, THIS JOURNAL, **38**, 400 (1916).
(9) R. D. Haworth and T. Richardson, *J. Chem. Soc.*, 348 (1936).

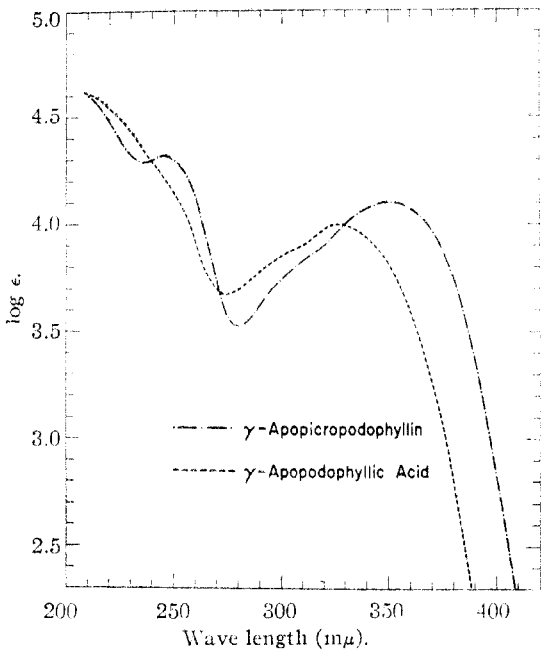
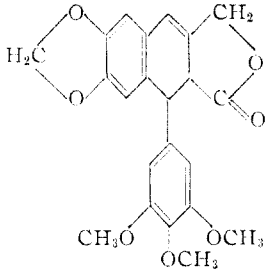


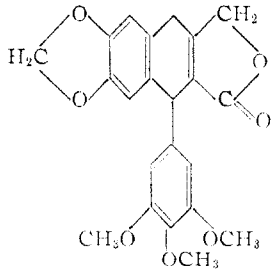
Fig. 3.—Ultraviolet absorption spectra in 95% ethanol.

that the optical activity was caused by the presence of structural isomers. Thus it appears that partially racemized γ-apopodophyllic acid was

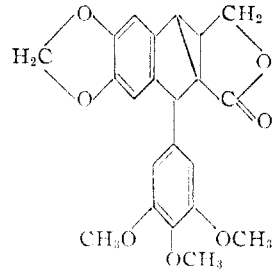


I

α-Apopicropodophyllin

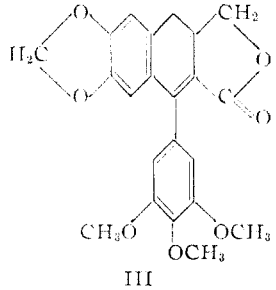


IIa



IIb

β-Apopicropodophyllin



III

γ-Apopicropodophyllin

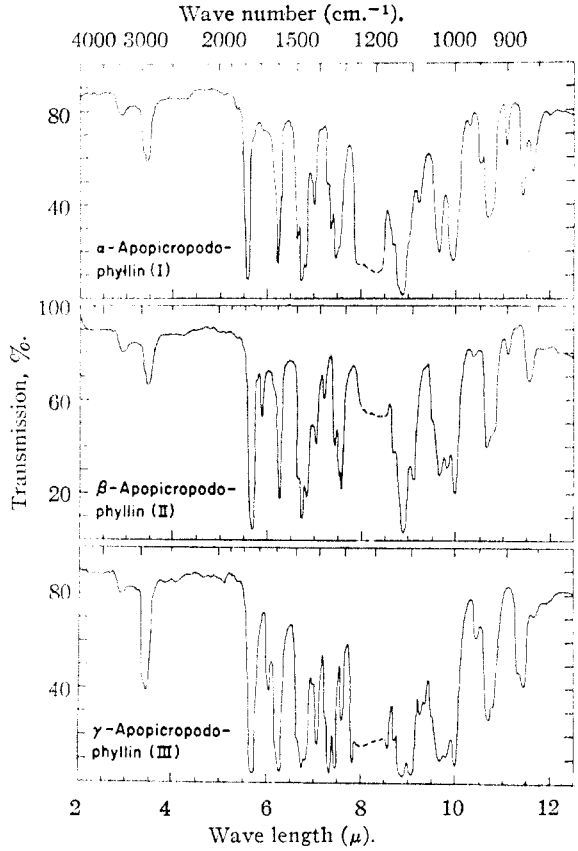


Fig. 4.—Infrared absorption spectra in chloroform.

formed in the base-catalyzed isomerization of α-apopodophyllic acid and that the more insoluble racemic acid crystallized first. Surprisingly, however, when pure γ-apopodophyllic acid was prepared from the dextrorotatory lactone, it was found to be devoid of any optical activity.

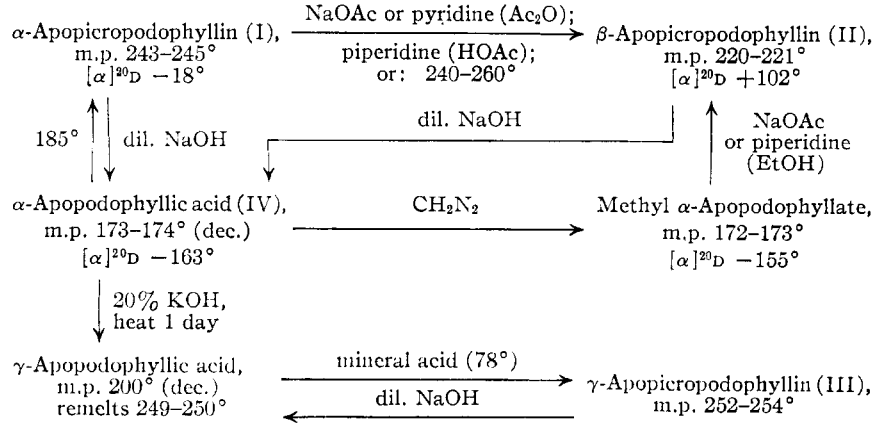
The interconversions relating the three apopicropodophyllins and the two apopodophyllic acids are summarized in Chart I.

Robertson and Waters[7] suggested that α- and β-apopicropodophyllin were geometrical isomers of a structure similar to III (except that the lactone ring was reversed); their proposed formula, apart from its structural incorrectness, had only one asymmetric carbon atom, and they apparently failed to notice the impossibility of ethylenic *cis–trans* isomerism in cyclohexene derivatives.

Chemical and spectroscopic evidence is now available which definitely establishes the structure of α-apopicropodophyllin as in I. In the case of β-apopicropodophyllin, IIa appears to be substantiated by the ultraviolet and infrared absorption spectra, to the exclusion of the alternate structure IIb (or some similar bicyclic formulation), which had been considered because the presence of an ethylenic double bond could not be detected by chemical means.

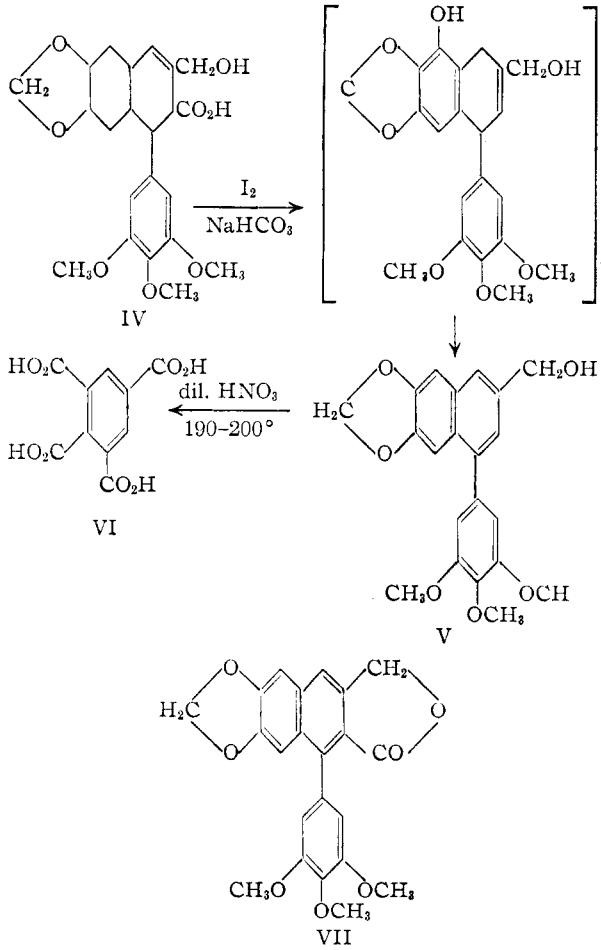
The fact, already mentioned, that α-apopicropodophyllin (and the corresponding hydroxy acid) decolorizes bromine rapidly, while β- and γ-apopicropodophyllin fail to do so, is consistent with the proposed structures. Only the double bond in I should be expected to add bromine readily, since

CHART I



the ethylenic linkages in IIa and III are completely substituted and rendered even more unreactive by the lactone carboxyl group.[10] Unsuccessful attempts were made to obtain a 1,2-glycol from α- or from β-apopicropodophyllin. It appears that even if such a glycol were formed as an intermediate, it would be dehydrated very readily to the completely aromatic dehydroanhydropicropodophyllin (VII).[6,9] This compound had previously been prepared from Haworth's synthetic γ-apopicropodophyllin by treatment with lead tetraacetate[9]; it has now been obtained from the α- and β-isomers in 77 and 58% yields, respectively, by the same method. Iodosilver benzoate, which has been used to prepare glycol benzoates from unsaturated compounds,[11] gave a 66% yield of dehydroanhydropicropodophyllin with α-apopicropodophyllin, but only a 10% yield with the β-isomer. Ozonization experiments, carried out with both α- and β-apopicropodophyllin, did not lead to the isolation of any pure substance in either case; mixtures of what appeared to be water-soluble polyfunctional compounds were obtained.

An oxidation reaction which conclusively established the structure of α-apopodophyllic acid (IV), and hence of the corresponding lactone (I), involved the use of iodine in bicarbonate solution. It has been shown that β,γ-unsaturated acids react readily with this reagent,[12,13] while α,β-unsaturated acids react very slowly[14] or not at all.[12] When the procedure was applied to γ-apopodophyllic acid, the starting material was recovered in 73% yield after 28 hours, while a small amount of an iodine-containing substance, insoluble in aqueous bicarbonate, was isolated, but not further studied. On the other hand, α-apopodophyllic acid (IV) underwent oxidative decarboxylation to 6,7-methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-hydroxymethylnaphthalene (V), m.p. 148.0–148.6°, which was obtained in 89% yield. This compound formed an acetate, m.p. 151.3–152.0°, and also a trinitrofluorenone complex,[15] m.p. 170–171.5°, characteristic for condensed aromatic ring systems. The structure of V is substantiated by the comparison[1] of its ultraviolet absorption spectrum with those of 6,7-methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-methylnaphthalene and of dehydroanhydropicropodophyllin (VII). Oxidation of V with dilute nitric acid at 190–200° in a sealed tube[16] gave benzene-1,2,3,5-tetracarboxylic acid (VI).[17] Formation of this acid rather than of the 1,2,3,4-isomer established both the carbon skeleton of V and the point of attachment of the hydroxymethyl group in IV and hence in the other derivatives of podophyllotoxin. The only positive evidence for the respective positions of the hydroxymethyl and the carboxyl groups had so far been Haworth's synthesis of VII,[9] which, however, included a step in which the carbon skeleton had under-



(10) C. F. H. Allen and A. H. Blatt in H. Gilman, "Organic Chemistry," Vol. I, 2nd ed., John Wiley and Sons, Inc., New York, N. Y., 1943, pp. 637, 683.

(11) Ch. Prévost, *Compt. rend.*, **196**, 1129 (1933); *cf.* ref. 10, p. 635.

(12) J. Bougault, *Ann. chim. phys.*, **14**, 145 (1908); **15**, 296 (1908); **22**, 125 (1911).

(13) A. W. Schrecker, G. Y. Greenberg and J. L. Hartwell, THIS JOURNAL, **74**, 5669 (1952).

(14) R. P. Linstead and C. J. May, *J. Chem. Soc.*, 2505 (1927).

(15) M. Orchin and E. O. Woolfolk, THIS JOURNAL, **68**, 1727 (1946).

(16) W. P. Campbell and D. Todd, *ibid.*, **62**, 1287 (1940).

(17) (a) L. I. Smith and G. D. Byrkit, *ibid.*, **55**, 4305 (1933); (b) D. E. Read and C. B. Purves, *ibid.*, **74**, 116 (1952).

gone rearrangement.[1] The formation of V from IV is analogous to that of 2,4,4-trimethylcyclohexenol from α-cyclogeranic acid[12,13]; the intermediate shown in brackets may be expected to be dehydrated readily to the stable naphthalene derivative.

On the basis of known reactions, the isomerizations summarized in Chart I support structure IIa for β-apopicropodophyllin. Many examples of base-catalyzed interconversions of α,β- and β,γ-unsaturated acids and esters have been described.[18] It may be pointed out that, if IIa is the correct formula for β-apopicropodophyllin, the tautomerization of the β,γ-unsaturated lactone (I) and methyl ester leads quantitatively to the α,β-unsaturated lactone (IIa), which in turn yields the β,γ-unsaturated acid (IV) exclusively when saponified. The α,β-unsaturated acid corresponding to III should be expected to be the most stable isomer of all since the double bond is conjugated with both the carboxyl group and the aromatic rings. It is obtained only under much more strenuous conditions, and it has already been mentioned that this is analogous to the formation of the stable 3,4-dihydro-2-naphthoic acid by drastic alkali treatment[8] of the 1,2- and 1,4-isomers.[19] The alternate formula IIb for β-apopicropodophyllin does not seem to be compatible with the isomerizations that have been described. It is true that there exist also examples for interconversions between unsaturated compounds and cyclopropane derivatives,[20] such as the *i*-steroid rearrangement[21] and somewhat similar reactions in the terpene field,[22] but the analogy of these reactions with any postulated interconversion between structure I and IIb (or a similar bicyclic formulation) appears farfetched.

Ultraviolet Spectra.—The postulated structures of α-apopicropodophyllin (I) and of the γ-isomer (III) are consistent with their ultraviolet absorption spectra, while IIa, only, and not IIb seems to be compatible with that of β-apopicropodophyllin. The absorption spectra of podophyllotoxin and of the three apopicropodophyllins are compared in Fig. 2. In β-apopicropodophyllin (IIa), the isolated α,β-unsaturated lactone grouping, which by itself would show intensive absorption only at shorter wave lengths,[23] should not be expected to influence greatly the main chromophoric system, and indeed the spectrum of this compound is rather similar to that of podophyllotoxin. If β-apopicropodophyllin had a bicyclic structure such as IIb, the conjugation between the cyclopropane ring and the aromatic nucleus should produce some shift of the absorption maximum toward the visible.[24] This maximum, however, is actually situated at a slightly shorter wave length in β-apopicropodophyllin (290 mμ) than in podophyllotoxin (292 mμ). Furthermore, Drake and Price[3] have found that the wave length of the absorption maximum remains unchanged when β-apopicropodophyllin is hydrogenated to the corresponding tetralin derivative, desoxypicropodophyllin. In α-apopicropodophyllin (I), on the other hand, the conjugation between the ethylenic double bond and the methylenedioxybenzene ring produces the expected shift of the absorption maximum toward the longer wave lengths. The position of this maximum (311 mμ) is only slightly changed (to 308 mμ) when the lactone ring is opened (Fig. 1), as may be expected in the case of an unconjugated carboxyl group. While the absorption spectra of α-apopodophyllic acid (IV) and of its methyl ester are almost identical, absorption at the minimum is reduced in the lactone. In γ-apopicropodophyllin (III), the conjugated system comprises at least the methylenedioxybenzene ring, the double bond and the lactone carboxyl group.[25] The number of possible charged resonance structures contributing to the excited states[26] is thus increased, and absorption is displaced so much toward the visible (max. 350 mμ) that solutions of the compound are yellow. The lesser conjugation effect of a carboxylic acid group as compared with that of an ester group is demonstrated by the somewhat smaller shift of the maximum (to 327 mμ) in γ-apopodophyllic acid (Fig. 3). The ultraviolet absorption spectra are thus in good agreement with what might be expected on theoretical grounds; additional substantiating evidence was obtained from a study of model compounds, namely, of the isomeric dihydro-β-naphthoic acids. It has been shown[13] that 1,4-dihydro-2-naphthoic acid has an absorption spectrum which appears to result roughly from the addition of the absorptions of the tetrahydro acid and of the α,β-unsaturated acid grouping; the shift toward the visible is most pronounced in the 3,4-dihydro acid, while the 1,2-isomer occupies a somewhat intermediate position. The analogy between the 1,4-dihydro acid and IIa, between the 1,2-dihydro acid and I, and between the 3,4-dihydro acid and III becomes clear when both the formulas and the absorption spectra are compared.

Infrared Spectra (Fig. 4).—While no peak that could be assigned to a cyclopropane ring[27] was detected in β-apopicropodophyllin, a band at 1698 cm.$^{-1}$ was present, which appears to correspond to a C=C stretching vibration, since the lactone carbonyl maximum is already accounted for by the peak located at 1760 cm.$^{-1}$. Double bond maxima at 1695 cm.$^{-1}$ have been observed in dihydroanisole derivatives[28] which are structurally similar to IIa. α-Apopicropodophyllin failed to show a maximum in the C=C stretching region, while such a peak was present in the γ-isomer (at 1648 cm.$^{-1}$). However, the absence of this band in I is not surprising, since it is usually very weak in the case of

(18) (a) G. A. R. Kon, *Ann. Repts. on Progress Chem.*, Chem. Soc. London), **29**, 136 (1932); (b) J. W. Baker, "Tautomerism," Routledge and Sons, London, 1934, p. 154 ff. *Cf.* also ref. 8.

(19) The only asymmetric carbon atom present in III is symmetric in both I and IIa. The optical activity of some of the samples of γ-apopicropodophyllin obtained from I by treatment with alkali and subsequent relactonization is therefore somewhat anomalous.

(20) *Cf.* ref. 18b, p. 175.

(21) L. F. Fieser and M. Fieser, "Natural Products Related to Phenanthrene," 3rd ed., Reinhold Publishing Corp., New York, N. Y., 1949, p. 256 ff.

(22) J. D. Roberts, *et al.*, THIS JOURNAL, **72**, 3116 (1950).

(23) L. J. Haynes and E. R. H. Jones, *J. Chem. Soc.*, 954 (1946).

(24) M. T. Rogers, THIS JOURNAL, **69**, 2544 (1947); *cf.* R. P. Mariella, *et al.*, *ibid.*, **70**, 1193 (1948); **74**, 518, 521 (1952); F. M. Klotz, *ibid.*, **66**, 88 (1944).

(25) The influence of the trimethoxyphenyl group is uncertain because of possible steric hindrance.

(26) R. N. Jones, THIS JOURNAL, **67**, 2127 (1945).

(27) M. L. Josien, N. Fuson and A. S. Cary, *ibid.*, **73**, 4445 (1951).

(28) G. Stork, *ibid.*, **74**, 768 (1952).

ethylenic double bonds which are not conjugated with carbonyl groups.[29] It may be noted that absorption in the C=C stretching region was found to be present in 1,4- and 3,4-, but not in 1,2-dihydro-2-naphthoic acid.[13]

To conclude, the present investigation has permitted the determination of the structure of α-apopicropodophyllin (I) and of the new γ-apopicropodophyllin (III). In addition, the weight of available evidence favors IIa for β-apopicropodophyllin.

Experimental[30,31]

α-Apopicropodophyllin (I).—The following modification of Robertson and Waters' procedure[7] gave the best results. A solution of 20 g. of picropodophyllin in 125 cc. of boiling acetic anhydride was cooled rapidly to 80°, 0.1 cc. of concd. sulfuric acid diluted with 5 cc. of acetic anhydride was added, and the purple mixture, from which a crystalline magma sooh separated, heated on the steam-bath for 7 minutes, then kept in the ice-box. The yield of colorless solid, m.p. 241–245°, obtained by filtering and washing with cold acetic acid, water and ethanol, was 17.8 g. (93%). Similar runs gave yields ranging from 78 to 86% and material which melted at least above 239°. Recrystallization from glacial acetic acid containing a trace of hydrochloric acid afforded colorless cottony needles, m.p. 243–245°, $[\alpha]^{20}D$ −18° (*c* 0.5, chloroform) [lit. m.p. 244–245°, $[\alpha]^{21}_{5461}$ −17.5° (*c* 0.49, chloroform),[7] m.p. 235.4–236.3°, $[\alpha]^{25}D$ −18° (*c* 0.581, chloroform)[3]]. When the material was recrystallized from organic solvents in the absence of mineral acid, a decrease in the melting point was often observed. The ultraviolet spectrum of the solution obtained by dissolving the pure compound in boiling ethanol indicated a more or less pronounced conversion to β-apopicropodophyllin, which, however, did not occur in the presence of hydrochloric acid.

β-Apopicropodophyllin (II). (a) With Pyridine in Acetic Anhydride.—Conversion of α-apopicropodophyllin by the method of Robertson and Waters[7] afforded a quantitative yield of crude material, m.p. 214–217°, which after two recrystallizations from ethyl acetate–ethanol gave colorless felt-like needles, m.p. 220.5–220.9°, $[\alpha]^{20}D$ +99° (*c* 0.5, chloroform) [lit.[7] m.p. 216°, $[\alpha]^{21}_{5461}$ +117.6° (*c* 0.51, chloroform)].

Anal. Calcd. for $C_{22}H_{20}O_7$: C, 66.66; H, 5.09. Found: C, 66.64; H, 5.00.

(b) With Piperidine in Acetic Acid.—This procedure has the advantage over the preceding one that a colorless material is obtained directly. A mixture of 1.2 g. of α-apopicropodophyllin, 12 cc. of glacial acetic acid and 0.12 cc. of piperidine was refluxed for one hour, and the solution gradually diluted with 120 cc. of water, then kept in the ice-box for 2 hours. The colorless felt-like needles (1.16 g., 97%) melted at 219–220.4°; recrystallization from chloroform–ethanol did not change the melting point; $[\alpha]^{21}D$ +103° (*c* 0.5, chloroform). A similar experiment gave, after two recrystallizations from chloroform–ethanol, material melting at 220–221°, $[\alpha]^{19}D$ +101° (*c* 0.5, chloroform).

(c) From Picropodophyllin Benzoate.—Pyrolysis of picropodophyllin benzoate, according to Drake and Price,[3] followed by crystallization from ethyl acetate, gave a 69.5% yield of material, m.p. 220–221° (lit.[3] 214.0–215.4°).

(d) By Heating of α-Apopicropodophyllin.—When 0.5 g. of α-apopicropodophyllin was heated at 260° until it had melted, then between 240 and 250° for 15 minutes, and the melt recrystallized from ethyl acetate, 0.375 g. of β-apopicropodophyllin, m.p. and mixed m.p. 219.5°, was obtained.

α-Apopodophyllic Acid (IV).—This acid was prepared by the method of Robertson and Waters[7] in 83 to 90% yield from either α- or β-apopicropodophyllin. It formed colorless needles, m.p. 173–174° (dec.) (lit.[7] 174°), after recrystallization from benzene, then from dilute ethanol. The value for the specific rotation were identical in both cases, but greatly different from those reported by Robertson and Waters: $[\alpha]^{20}D$ −163° (*c* 1, chloroform) [lit.[7] $[\alpha]^{21}_{5461}$ −279.4° (*c* 1, chloroform)]. The acid was recovered unchanged (m.p. 173–174° dec., $[\alpha]^{20}D$ −161°, after recrystallization) when refluxed with sodium acetate in ethanol for 2.5 hours.

Methyl α-Apopodophyllate.—A distilled ethereal solution (69 cc.) of diazomethane (from 6.9 g. of nitrosomethylurea) was added with external cooling to a solution of 4.6 g. of IV in 23 cc. of methanol, and the mixture kept in the ice-box for 2.5 hours. The yellowish prisms, which gradually separated, were collected and washed with absolute ether; the yield of material, m.p. 167–173, was 3.83 g. Another 0.70 g. was obtained from the mother liquor by evaporation and recrystallization, bringing the total yield to 95%. Recrystallization from methanol or, preferably, from chloroform–hexane afforded colorless prisms; m.p. 172–173° (some variation with the heating rate), $[\alpha]^{20}D$ −155° (*c* 1, chloroform). The substance gave a purple color with concd. sulfuric acid, and its chloroform solution decolorized bromine rapidly.

Anal. Calcd. for $C_{23}H_{24}O_8$: C, 64.48; H, 5.65; 4 OCH_3, 28.98. Found: C, 64.69; H, 5.89; OCH_3, 28.85.

The compound was recovered unchanged in 97% yield when heated in pyridine solution for one hour on the steam-bath. β-Apopicropodophyllin (II), m.p. 217–220° (219–220° after recrystallization; no depression with an authentic sample), was isolated in 86% yield when the ester was refluxed with sodium acetate in 80% ethanol for 1.5 hr., and in 97% yield when it was boiled with piperidine in 95% ethanol for one hour. Similarly, β-apopicropodophyllin (m.p. and mixed m.p. 220–221°, $[\alpha]^{20}D$ +105°, after recrystallization) was obtained in 76% yield in a preliminary experiment when the ethereal diazomethane solution employed in the methylation of IV had not been distilled and the reaction mixture was allowed to stand at room temperature for 20 hours.

γ-Apopodophyllic Acid.—A solution of 17 g. of α-apopicropodophyllin in 320 g. of 20% aqueous potassium hydroxide was refluxed in a stainless steel flask under oxygen-free nitrogen for 24 hours. The red-brown solution was cooled in ice, and excess free alkali neutralized with hydrochloric acid; then 7.2 g. of sodium bicarbonate was added, followed by a solution of 21.8 g. of iodine and 43.6 g. of potassium iodide in 80 cc. of water. After 16 hours, the black solid,[32] which had separated rapidly with evolution of carbon dioxide, was removed by filtration and washed with water. Sulfur dioxide was passed through the filtrate with external cooling until disappearance of the iodine color. The mixture, which was acid to congo red and from which a yellow precipitate had separated, was extracted with about 200 cc. of chloroform, and the extract washed twice with water, then dried over sodium sulfate. The oily residue remaining after evaporation of the chloroform was dissolved in hot benzene. A pale-yellow cottony solid separated upon cooling; yield 0.67 g. (3.8%).[33] The crude material melted with foaming when immersed at 200°, resolidified, and melted again at 243–246°. Fractional crystallization from methanol–benzene gave 0.33 g. of colorless (first crop) and 0.29 g. of yellowish material (second crop), melting (after resolidification) at 247–250° and 236–248°, respectively.

The pure γ-apopodophyllic acid was obtained as small colorless needles by saponification with aqueous alcoholic sodium hydroxide of the γ-apopicropodophyllin prepared from these crude acid fractions, followed by acidification at 0°, isolation with chloroform, crystallization from benzene, and recrystallization from 30% ethanol. It melted at 249–

(29) R. N. Jones, P. Humphries, E. Packard and K. Dobriner, THE JOURNAL, **72**, 86 (1950).

(30) Melting points are corrected and were determined with the Hershberg apparatus.

(31) The ultraviolet absorption spectra were determined with a Beckman model DU spectrophotometer. The infrared spectra were measured with a Perkin–Elmer model 21 spectrometer; chloroform solutions were employed, and the absorption of that solvent between 1180 and 1250 cm.$^{-1}$ has masked any characteristic bands possibly present in that range.

(32) This by-product was identified as 6,7-methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-hydroxymethylnaphthalene (V), which after purification was obtained in 51% yield and had evidently been formed from unchanged α-apopodophyllic acid.

(33) Similar preparations gave yields reaching 4.7%. Attempts to increase the yield by raising the concentration of potassium hydroxide failed because of the insolubility of the potassium salt of α-apopodophyllic acid in the more concentrated solution. When the isomerization was attempted by treatment with aqueous barium hydroxide (*cf.* ref. 8) in a sealed tube at 160°, excessive tar formation took place and no γ-apopodophyllic acid could be isolated.

250° when immersed at room temperature, while it melted with foaming at 202° when immersed at 200°, resolidified and remelted at 249–250°. This behavior and the ultraviolet absorption spectrum duplicated those of 6,7-methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-hydroxymethyl-3,4-dihydro-2-naphthoic acid, prepared[1] by total synthesis.

Anal. Calcd. for $C_{22}H_{22}O_8$: C, 63.76; H, 5.35. Found: C, 63.61; H, 5.52.

γ-Apopicropodophyllin (III).—The separate fractions of crude γ-apopodophyllic acid were dissolved in 5 cc. of hot ethanol, and the solutions heated on the steam-bath for 20 minutes, after addition of 10 cc. of *N* hydrochloric acid.[34] The crystalline material, which separated rapidly, was dissolved in chloroform, chromatographed on alumina[35] and the yellow zone eluted with chloroform. Concentration with addition of ethanol furnished apparently colorless needles. The lactone obtained from the first crop of γ-apopodophyllic acid had m.p. 252–254°[36] and $[\alpha]^{19}D$ +0.1° (*c* 1, chloroform), while the material prepared from the second crop had m.p. 250–254° and $[\alpha]^{19}D$ +27.8° (*c* 1, chloroform). The γ-apopodophyllic acid formed from both the inactive and dextrorotatory lactones, however, proved to be entirely devoid of optical activity (in ethanol). γ-Apopicropodophyllin obtained in similar experiments had m.p. 252–253°, $[\alpha]^{21}D$ +26.4° (*c* 0.5, chloroform); m.p. 253–254°, $[\alpha]^{23}D$ +3.4° (*c* 0.8, chloroform); m.p. 248–254°, $[\alpha]^{19}D$ +25.0° (*c* 1, chloroform). Both the optically active and inactive samples had identical ultraviolet absorption spectra, which were furthermore indistinguishable from that of the synthetic lactone[1]; thus the presence of structural isomers appears to be excluded. The infrared spectra in chloroform of the material, $[\alpha]D$ +26.4°, and of the synthetic lactone were also identical.

Anal. Calcd. for $C_{22}H_{20}O_7$: C, 66.66; H, 5.09. Found: C, 66.70; H, 5.11.

Dehydroanhydropicropodophyllin (VII). (a) **From α-Apopicropodophyllin with Lead Tetraacetate.**—A suspension of 0.6 g. of α-apopicropodophyllin and 1.2 g. of lead tetraacetate in 15 cc. of glacial acetic acid was heated at 75° for 15 minutes. A yellow solution formed at first from which crystals soon separated. The mixture was diluted with water, extracted with chloroform, and the extract washed with water, aqueous sodium bicarbonate and again water, dried over sodium sulfate and concentrated with addition of ethanol. The yield of colorless small needles, m.p. 268–269° (lit.[9] 267–268°), was 0.46 g. (77%). The compound did not depress the melting point of a sample, m.p. 270–271° (lit.[6] 266°), prepared by dehydrogenation of picropodophyllin with palladium[6] at 240°, followed by three recrystallizations from chloroform–ethanol. Its solutions showed a bright blue fluorescence under ultraviolet light. Attempts to prepare a trinitrofluorenone complex[15] were unsuccessful.

(b) **From β-Apopicropodophyllin with Lead Tetraacetate.**—By the same procedure, with the exception that the dark chloroform solution was chromatographed over alumina, a 58% yield of dehydroanhydropicropodophyllin was obtained.

(c) **From α-Apopicropodophyllin with Iodosilver Benzoate.**[11]—To a suspension of 1.53 g. of silver benzoate in 9 cc. of anhydrous benzene was added with shaking a solution of 0.635 g. of iodine in 8 cc. of benzene, then, as soon as the iodine color had disappeared, a solution of 0.99 g. of α-apopicropodophyllin in 20 cc. of hot ethylene chloride. The mixture was refluxed with exclusion of moisture for 24 hours, then silver iodide was removed by filtration and washed with hot chloroform. The combined light yellow filtrate and washings were evaporated to dryness and the residue triturated with hexane. The crystalline material (1.19 g.), which melted over a wide range, gave after recrystallization from chloroform–ethanol (Norit) 0.65 g. (66%) of colorless needles, m.p. and mixed m.p. 270–271°.

(d) **From β-Apopicropodophyllin with Iodosilver Benzoate.**—A mixture of 0.99 g. of β-apopicropodophyllin, 1.15 g. of silver benzoate, 0.635 g. of iodine and 50 cc. of toluene was refluxed for 24 hours, and silver iodide removed by filtration. Addition of hexane to the filtrate gave 0.74 g. of a low-melting solid, which after recrystallization from chloroform–ethanol, then from ethyl acetate, yielded 0.10 g. (10%) of dehydroanhydropicropodophyllin, m.p. 264–266°. No pure compound could be isolated from the mother liquors.

6,7-Methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-hydroxymethylnaphthalene (V).—Solutions of 2.5 g. of α-apopodophyllic acid (IV) in 19.5 cc. of *N* sodium bicarbonate and of 3.125 g. of iodine and 6.3 g. of potassium iodide in 20 cc. of water were mixed and kept at room temperature in the dark for 19 hours. The black solid, which had started to separate almost immediately with evolution of carbon dioxide, was collected and dissolved in chloroform. The brown solution was decolorized by shaking with aqueous sodium thiosulfate, washed with sodium bicarbonate solution, then with water, dried over sodium sulfate and concentrated. Addition of hexane to incipient cloudiness afforded 1.80 g. of colorless crystals, m.p. 148.0–148.6°. A second crop of 0.18 g., m.p. 135–142°, was obtained from the mother liquor, bringing the total yield to 89%. Crystallization of the first crop from benzene–hexane gave felt-like clusters of small needles without an increase in the melting point.

Anal. Calcd. for $C_{21}H_{20}O_6$: C, 68.47; H, 5.47. Found: C, 68.37; H, 5.65.

The acetyl derivative was prepared in 97% yield by refluxing 0.25 g. of the material with 0.25 g. of anhydrous sodium acetate and 5 cc. of acetic anhydride for 2.5 hours. The crude product, obtained by diluting with water, melted at 149–151°. Crystallization from ethanol gave small colorless needles, m.p. 151.3–152.0°.

Anal. Calcd. for $C_{23}H_{22}O_7$: C, 67.31; H, 5.40. Found: C, 67.27; H, 5.47.

The trinitrofluorenone complex[15] was prepared by dissolving 50 mg. of the hydroxymethyl compound and 42 mg. of trinitrofluorenone in hot benzene. Addition of an equal volume of hexane, and scratching the walls of the vessel, caused separation of 64 mg. of crystals, m.p. 169.9–171.3°. Recrystallization from benzene–hexane gave small dark purplish-red prisms, m.p. 170.0–171.5°. Attempts to prepare or to recrystallize this complex in the presence of ethanol proved unsuccessful; apparently the addition compound is more soluble in that solvent than trinitrofluorenone itself.

Anal. Calcd. for $2C_{21}H_{20}O_6 \cdot C_{13}H_5O_7N_3$: C, 62.79; H, 4.31; N, 3.99. Found: C, 62.54, H, 4.33; N, 3.76.

When 50 mg. of γ-apopodophyllic acid was treated with iodine in bicarbonate solution in the proportions that had been used in the case of the α-isomer (IV), a dark brown solid separated slowly. It was collected after 28 hours and washed with dilute sodium bicarbonate, then triturated with aqueous sulfur dioxide to give 9 mg. of colorless material which melted at 162.5–163° with liberation of iodine and which was very soluble in most organic solvents. When the mother liquor was acidified with sulfur dioxide, a colorless solid separated. The suspension was boiled for several minutes, after addition of some hydrochloric acid, and 35 mg. (73%) of impure γ-apopicropodophyllin (III), m.p. 238–246°, was thus isolated. Recrystallization from chloroform–ethanol gave colorless needles, m.p. and mixed m.p. 248–250°.

Benzene-1,2,3,5-tetracarboxylic Acid (VI).—A mixture of 552 mg. of the hydroxymethylnaphthalene V and 2.9 cc. of 29% nitric acid was heated at 190–200° in a sealed tube[16] for 27 hours. The tube was opened, resealed after addition of 1 cc. of concd. nitric acid, and heated for another 11 hours. The yellow solution was diluted with water, filtered and evaporated to dryness. Fuming nitric acid was added, and the mixture again evaporated to dryness. The crystalline residue was stirred with a small amount of fuming nitric acid, collected in a sintered-glass funnel, and washed with fuming nitric acid. The yellow material weighed 117 mg. (31%) and melted at 247–254° (foaming) [lit. 238–253°[17a]; 236–253° (crude hydrated), 243–247° (pure anhydrous)[17b]].

The tetramethyl ester, prepared with diazomethane, formed colorless fan-shaped aggregates of needles (from methanol); m.p. 109.6–110.2° [lit. 110.5–111°[16], 107–109°[17a], 111–116°[17b]], no m.p. depression with an authentic sample.[37]

Anal. Calcd. for $C_{14}H_{14}O_8$: C, 54.19; H, 4.55; OCH_3, 40.01. Found: C, 54.26; H, 4.75; OCH_3, 41.02 (small sample).

(34) In other experiments, lactonization was accomplished by vacuum sublimation (255° bath, 0.7 mm.).

(35) Alcoa Activated Alumina, grade F-20.

(36) No mixed melting point depression with synthetic (ref. 1) 6,7-methylenedioxy-1-(3,4,5-trimethoxyphenyl)-3-hydroxymethyl-3,4-dihydro-2-naphthoic acid lactone, m.p. 250–251°.

(37) Kindly furnished by Dr. Henry C. Howard; *cf.* G. Schulz and H. C. Howard, THIS JOURNAL, **68**, 991 (1946).

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[CONTRIBUTION FROM HICKRILL CHEMICAL RESEARCH LABORATORY]

Rearrangement of Halotropones. Chloride Exchange in Tribromotropolone

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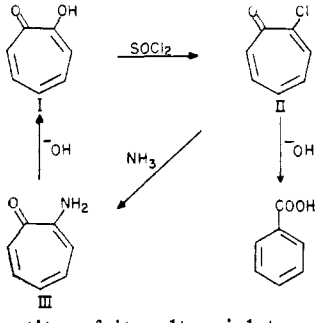
Chlorotropone, obtained from tropolone and thionyl chloride, reacts with ammonia normally but slowly to give aminotropone whereas with hydroxide ion benzoic acid is produced rapidly, this being a very facile rearrangement to the aromatic system. Upon treatment with sodium methoxide, 2-methoxy-3,5,7-tribromotropone rearranges to methyl 2,4,6-tribromobenzoate and 2-methoxy-3,5-dibromobenzoic acid. A similar behavior is shown by 2-methoxy-3,5,7-trichlorotropone while 2,3,5,7-tetrachlorotropone rearranges with alkali to 2,3,5-trichlorobenzoic acid. 3,5,7-Tribromotropolone reacts with thionyl chloride to give 2,3,5,7-tetrachlorotropone and with concentrated hydrochloric acid to give 3,5,7-trichlorotropolone. This unusual, acid-catalyzed halide exchange is a unique consequence of the cyclic nature of tropolone and has occasioned the reexamination of the theoretical aspects of the aromatic character of tropolone.

The hydroxyl function of tropolone (I), which simulates the hydroxyl of a carboxylic acid in its acidity and etherification with methanolic hydrogen chloride and an alcohol or phenol in its conversions to an acetate and a benzoate,[2] reacts with thionyl chloride to give 2-chlorotropone (II). The structure of II is firmly established by its conversion to aminotropone (III)[2] with ammonia and by the



essential identity of its ultraviolet spectrum with that of tropolone methyl ether (IV) (Fig. 1).

Hydroxide ion, which hydrolyzes III, IV and tropolone acetate to tropolone, reacts with chlorotropone rapidly at 100° to give benzoate ion, no trace of tropolone being detectable. This facile, aromatizing rearrangement of the tropolone system is to be added to an increasingly large group: the benzilic acid type which is difficult with tropolone[2] but which is facilitated by nitro groups[3]; the methoxide-catalyzed rearrangement of tropolone methyl ether to methyl benzoate[2]; the iodinative decarboxylation of tropolone to triiodophenol[2,4]; and the deaminative aromatization of α-aminotropolones to salicylic acids.[6,7]

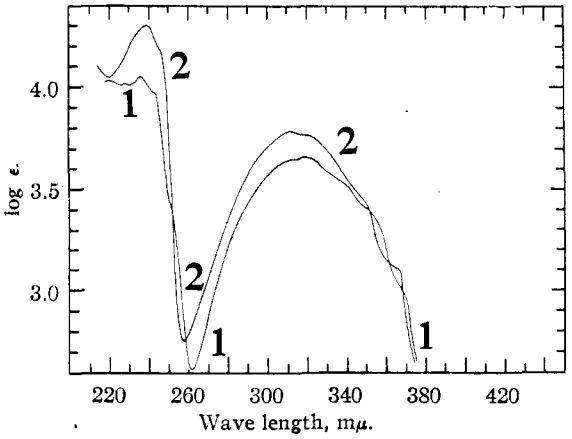


Fig. 1.—Ultraviolet absorption spectra of tropolone methyl ether (IV, curve 1) and of chlorotropone (II, curve 2) in isoöctane.

As a more complicated example, 3,5,7-tribromotropolone (V) as its methyl ether (VI) was treated with methoxide ion in boiling, absolute methanol. The major product, as recently reported by Cook, *et al.*,[5] in support of the structure originally proposed[2] for V, is indeed methyl 2,4,6-tribromobenzoate (VII), but a minor product, 3,5-dibromo-2-methoxybenzoic acid (VIII), is produced in significant quantity. That the appearance of VIII as the free acid and not as the expected methyl ester (IX) results from subsequent reaction of IX with methoxide ion is indicated by the fact that methyl 3,5-dibromo-2-methoxybenzoate (IX) reacts with absolute methanolic sodium methoxide to give the acid (VIII) and (presumably) dimethyl ether.[8] In a similar fashion 3,5,7-trichlorotropolone methyl

(1) Department of Chemistry, Yale University, New Haven, Conn.

(2) W. von E. Doering and L. H. Knox, THIS JOURNAL, **73**, 828 (1951).

(3) An effect first discovered by T. Nozoe, Y. Kitahara, K. Yamane and K. Yamaki, *Proc. Japan Acad.*, **26**, No. 8, 14 (1950), in the rapid rearrangement of 3,5-dinitro-6-isopropyltropolone.

(4) In their first paper on tropolone, Cook, *et al.* [J. W. Cook, A. R. Gibb, R. A. Raphael and A. R. Somerville, *J. Chem. Soc.*, 503 (1951)] maintained that these three rearrangements could not be realized, a position which they reversed in their third.[5]

(5) J. W. Cook, A. R. M. Gibb and R. A. Raphael, *ibid.*, 2244 (1951).

(6) R. D. Haworth and P. R. Jeffries, *ibid.*, 2067 (1951).

(7) T. Nozoe, Y. Kitahara and K. Doi, THIS JOURNAL, **73**, 1895 (1951).

(8) See, for examples, J. F. Bunnett, M. M. Robison and F. C. Pennington, *ibid.*, **72**, 2378 (1950).