acid was also characterized as a *p*-bromophenacyl ester, prepared as described by Shriner, Fuson, and Curtin.[21] A 0.091-g. portion of the acid yielded 0.043 g. of *p*-bromophenacyl ester, m.p. 129–131°, undepressed on admixture with authentic *p*-bromophenacyl nonandioate. The volatile acid (0.101 g.) was similarly converted to a *p*-bromophenacyl ester, 0.011 g. after two recrystallizations from 50% ethanol, m.p. 68–70°, undepressed on admixture with authentic *p*-bromophenacyl hexanoate. The malonic acid fragment (VII) was not isolated.

Acknowledgment. The authors wish to thank Mrs. Clara McGrew for microanalyses; Dr. E. J. Dufek for furnishing a sample of *threo*-9,10-dihydroxyoctadecanoic acid; and Dr. Quentin Jones, Crops Research Division, Agricultural Research Service, U. S. Department of Agriculture, for his cooperation in obtaining seeds.

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Some Neutral Components of Cigarette Smoke[1]

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Received September 11, 1959

The paraffin wax fraction of cigarette smoke has been shown to contain the fifteen normal alkanes from docosane to hexatriacontane and branched alkanes having between 21 and 32 carbon atoms, inclusive. About one third of this paraffin mixture consists of hentriacontane and tritriacontane. Also present in the neutral fraction of smoke are squalene, isosqualene, stigmasterol, β-sitosterol, and probably γ-sitosterol.

Carcinogenicity tests on mice[3] of fractions of cigarette smoke indicated that the fractions designated as K and M[4] were the most active. We have previously described, in part, the chemical composition of these materials; the present paper reports our further studies on the neutral components of M and related fractions.

An earlier paper from our laboratory[4] had presented indicative proof of the presence of hentriacontane and tritriacontane in the smoke of blended American cigarettes. A later report by Cuzin *et. al.*[5] stated that no evidence was found for the presence in smoke (or tobacco leaf) of alkanes of more than 32 carbons.[6] A reinvestigation of our material was accordingly undertaken. A purified paraffin mixture of M'[4] was prepared as before and its x-ray diffractogram was compared with those of synthetic samples of hentriacontane, tritriacontane, and pentriacontane; the respective values for the long-spacings were 42.7 Å, 41.8 Å, 44.1 Å, and 46.7 Å.[7] These results confirmed those obtained earlier by us in that the value for the wax from smoke fell between those of the C_{31} and C_{33} hydrocarbons; we were later able to obtain a mass spectrometric analysis which clearly showed this wax to be composed largely of equal amounts of the C_{31} and C_{33} *normal* alkanes, some C_{32} homolog, and small amounts of other alkanes (Table I).

TABLE I

MASS SPECTRA OF PARAFFIN MIXTURE FROM M'

No. of Carbons	% Normal	% Branched
30	2	
31	38	1
32	14	
33	39	1
34	5	

To obtain a broader analysis of the higher-alkane distribution in cigarette smoke, a sample of MM[4] was exhaustively extracted with concentrated

(1) Portions of this paper were presented at the Seventh International Cancer Congress, London, July 1958 and at the Meeting-in-Miniature of the New York Section of the American Chemical Society, April 1959; *cf.* also A. I. Kosak and J. S. Swinehart, *Chem. and Ind. (London)*, 1007 (1958).

(2) Abstracted from a part of the Ph.D. thesis of J.S.S., New York University, April 1959.

(3) W. E. Smith, N. Nelson, L. Orris, and A. I. Kosak, unpublished data.

(4) A. I. Kosak, J. S. Swinehart, and D. Taber, *J. Natl. Cancer Inst.*, **17**, 375 (1956).

(5) J. L. Cuzin, L. V. Thoi, and M. S. Morec, paper presented at the second International Tobacco Science Congress, Brussels, June 1958.

(6) The x-ray diffraction data listed by these workers for their highest molecular weight fraction was the same as that which we had reported and the difference is one of interpretation. They ascribed their *d*-value of 42.7 Å to dotriacontane. We felt that the *d*-value of this magnitude more probably represented a mixture of *odd*-numbered alkanes for four reasons: first, the paraffins of other plant waxes consist principally of this class of alkanes; second, a mixture of normal alkanes will have the same long-spacing as that of a single hydrocarbon whose molecular weight equals that of the average of the mixture, if the hydrocarbons of the mixture do not differ from each other by more than four carbon atoms [S. H. Piper, A. C. Chibnall, S. J. Hopkins, A. Pollard, J. A. B. Smith, and E. F. Williams, *Biochem. J.*, **25**, 2072 (1931)]; third, the "blurred" diffraction lines (Piper *et al.*, *loc. cit.*); fourth, evidence for the occurrence of tritriacontane in tobacco leaf [A. C. Chibnall, S. H. Piper, A. Pollard, E. F. Williams, and P. N. Sahai, *Biochem. J.*, **28**, 2189 (1934)].

(7) The values for the synthetic materials are in good agreement with those reported by D. R. Kreger in J. Bouman, *Selected Topics in X-Ray Crystallography*, Amsterdam, 1951, p. 316.

TABLE II

PARAFFIN CONTENT OF MM

No. Carbons	Fraction 1, %		Fraction 2, %		Fraction 3, %		Fraction 4, %		Fraction 5, %		% of Total Alkanes in MM[a]	
	Normal	Branched	Normal	Branched	Normal	Branched	Normal	Branched	Normal	Branched	Normal	Branched
21		3										0.16
22	2	4									0.11	0.21
23	9	4									0.49	0.21
24	17	4									0.92	0.21
25	18	3	2								2.5	0.16
26	11	2	1	2							1.4	1.63
27	9	2	8	1							6.6	0.87
28	6	1	2	18		1		1			1.8	13.9
29	5		8	3				1			6.3	2.31
30			4	19	1	7	3	3	1	1	3.3	15.33
31			26	1	24	1	9	1	5		22.9	0.90
32			4	1	13	3	4		4	1	4.8	1.13
33					46		71		53		9.7	
34					4		7		13		1.2	
35									21		0.97	
36									1		0.05	
% of total paraffins	4.2	1.2	41.7	34.2	9.6	1.3	3.0	0.2	4.5	0.1	63.0	37.0

[a] These values are approximate since Fraction 1 contained 10–20% olefinic or naphthenic material of formula C_{n-2} with discrete peaks at C_{20} (m/e = 278) and C_{24} (m/e = 334) and Fraction 5 contained 2–3% of C_nH_{2n-6} material with peaks at C_{26}, C_{25}, and C_{40}.

sulfuric acid and was then separated by chromatography on alumina into five fractions which were also examined in the mass spectrometer. Table II indicates that hentriacontane is the principal component, and that considerable quantities of branched-chain alkanes are also present. Somewhat similar distributions of alkanes have been found in other plant waxes, marine sediments, and soil extracts.[8,9]

After removal of the paraffin hydrocarbons from a sample of fraction M[4] by chromatography on silica gel, a viscous oil was eluted whose infrared spectrum indicated it to be a mixture of polyolefins and long-chain ketones. Most of the ketonic material was removed by further chromatography on acid-washed and on basic alumina, and subsequent distillation yielded a cut boiling at 215–221°/1 mm., constituting about 0.02% of the total smoke condensate, whose infrared spectrum was that of squalene plus bands at the 6.07μ (w) and 11.25μ (m) regions where isosqualene absorbs strongly.[10] This material was hydrogenated over 5% rhodium on alumina catalyst and absorbed 6.1 mol. of hydrogen; the value expected for squalene is 6.0 mol. Upon treatment of the smoke component with hydrogen chloride in anhydrous acetone a hexahydrochloride was formed[11] which was identical with that produced from a freshly distilled sample of squalene under the same conditions. Identity was established by mixture melting point, infrared absorption, and x-ray diffraction pattern.

The presence of isoqualene was confirmed by chromatographing on paper the mixture isolated from smoke, "regenerated squalene" (which contains isosqualene) obtained from squalene hexahydrochloride,[10] and squalene. The results are presented in Table III. The isosqualene spot derived from the smoke component (R_f 0.82) was decidedly weaker than the squalene spot (R_f 0.68) from the same source, which is consistent with the spectroscopic data.

TABLE III

PAPER CHROMATOGRAPHY OF SQUALENE SAMPLES

Material	R_f Value
Squalene[a]	0.70
Regenerated squalene[b]	0.68, 0.84
Material from smoke[b]	0.64,[c] 0.69, 0.82, 0.91[c]
Acid-treated squalene[d]	0.69

[a] Average of four runs. [b] Three runs. [c] These spots were weak and diffuse. [d] One run.

(8) *E.g.* G. Wanless, W. King, and J. J. Ritter, *Biochem. J.*, **59**, 687 (1955); E. D. Evans, G. S. Kenny, M. G. Meinschein, and E. E. Bray, *Anal. Chem.*, **29**, 1859 (1957); W. E. Meinschein and G. S. Kenny, *Anal. Chem.*, **29**, 1153 (1957).

(9) Dr. W. Carruthers has informed us that he and Dr. R. A. W. Johnston have also detected branched-chain alkanes in cigarette smoke as well as in green and fermented tobacco leaves. We are indebted to Dr. Carruthers for making his results available to us before publication.

(10) W. G. Dauben, H. C. Bradlow, N. K. Freeman, D. Kritchevsky, and M. Kirk, *J. Am. Chem. Soc.*, **74**, 4321 (1952).

(11) I. M. Heilbron, E. Kamm, and W. M. Owens, *J. Chem. Soc.*, 1631 (1926).

No changes were noted in the infrared spectrum of a sample of squalene which had been agitated with 12% hydrochloric acid for 12 hr. nor did any new paper chromatographic spot appear. This precludes the possibility that the isosqualene found was an artifact produced during the acid extraction step employed in the primary fractionation[4] (see Table III).

Three steroids, stigmasterol,[12-14] β-sitosterol,[13] and γ-sitosterol,[13] have been reported to be in cigarette smoke. It is extremely difficult to isolate *pure* individual components from small amounts of mixtures of related sterols whose melting points and rotations are in the same range,[15] *e.g.*, the sitosterols. Proof of identity of such phytosterols is often complicated by the fact that C_{27}–C_{29} sterols with the same number of double bonds, identical oxygen functions, and similar but not identical structures often have identical infrared spectra.[16] We have isolated two steroids from fraction M in addition to the previously reported stigmasterol.[12] By a comparison of the melting points, rotations, and infrared spectra of these compounds and their esters with those of β- and γ-sitosterol, we have assigned the latter two structures to the smoke sterols, confirming the work of Carruthers and Johnston.[13] We have obtained what we believe to be positive evidence for the identities of β-sitosterol and stigmasterol by x-ray diffraction powder techniques[17] which showed the patterns of the isolated sterols to be identical with those of the respective authentic samples.

EXPERIMENTAL[18]

15-Tritriacontanone was synthesized in 72% yield from 1-bromohexadecane and margaroyl chloride *via* the organocadmium intermediate[4]; m.p. 73.4–76.8°. Chromatography on alumina and two recrystallizations from ethanol raised the m.p. to 79.7–81.7° [lit.[19] m.p. 78.8–79.2°].

18-Pentatriacontanone was prepared by the method of Kipping[20] in 40% yield; m.p. 88.5–89.4° [lit.[21] m.p. 88.7–89.0°].

Tritriacontane and pentatriacontane. A mixture of 1.1 g. of ketone (15-tritriacontanone or 18-pentatriacontanone), 17 ml. of 70% hydrazine hydrate, and 65 ml. of diethylene glycol was maintained at 110° for 1 hr.; 6.5 g. of potassium hydroxide was then added and the mixture was heated at reflux for 8 hr. The mixture was cooled to 110° and the above steps were repeated four times with the addition of 10 ml. of hydrazine hydrate and 5 g. of potassium hydroxide in each stage; after the last addition, the reaction was heated at reflux for 72 hr.[22] The acidified reaction product was extracted with three 200-ml. portions of benzene, the solvent evaporated, and the residue was recrystallized from ethanol to give tritriacontane (30%), m.p. 71.4–71.7° [lit.[23] m.p. 71.5–71.6°] and pentatriacontane (26%), m.p. 74.2–74.6° [lit.[24] m.p. 74.4–74.5°].

X-Ray diffraction data for paraffins. The diffraction maxima (angles in degrees) were: paraffin wax from smoke,[25] 1.05, 2.07, 3.11, 4.13, 5.15, 6.07; hentriacontane[4] 1.06, 2.10, 3.16, 4.24, 5.29; tritriacontane: 2.95, 4.03, 5.04, 6.04, 7.03; pentatriacontane: 1.87, 2.85, 3.82, 4.77, 5.74, 6.70, 7.64, 8.56. Substitution of these values in Bragg's equation gave respective average values for *d* of 42.7 Å, 41.8 Å, 44.1 Å, 46.5 Å.

Fractionation of MM waxes for mass spectrographic study. A solution of 16 g. of MM in 140 ml. of petroleum ether (30–60°) was extracted successively with thirty 50-ml. portions of concentrated sulfuric acid, two 6-ml. portions of 5% aqueous sodium carbonate, and three 60-ml. portions of water. The residue was chromatographed on 5.5 kg. of basic alumina, and petroleum ether (30–60°) was passed through the column. Fraction 1 (0.6 g.), fraction 2 (8.4 g.), m.p. 50.0–61.0°, fraction 3 (1.2 g.), m.p. 58.0–67.5°; fraction 4 (0.38 g.), m.p. 62.0–68.9°; fraction 5, eluted with benzene, (0.51 g.), m.p. 64.0–70.3°.

Isolation of squalene. In a typical run, 25 g. of fraction M was chromatographed on 2800 g. of silica gel. The column was eluted successively with 30–60° petroleum ether (1.6 l.), 1:4 benzene–petroleum ether (2.3 l.), and 2:3 benzene–petroleum ether (2.8 l.). Evaporation of the solvent from the last eluate left 5 g. of viscous, orange oil, 2.2 g. of which was chromatographed on 1800 g. of Merck acid washed alumina and eluted with 2 l. of petroleum ether. The product thus obtained from several runs (9.3 g.) was put through a column of 800 g. of basic alumina and the petroleum ether eluate (300 ml.) consisted of 4.2 g. of light yellow oil which still showed some infrared absorption in the carbonyl region. It was distilled and gave 0.31 g. of colorless oil, b.p. 215–221°/1 mm., whose infrared spectrum resembled that of squalene with the addition of peaks at 6.07 μ and 11.25 μ.

Squalene hexahydrochloride was prepared by the method of Heilbron *et al.*[11] both from the isolated squalene mixture (m.p. 109.5–112.0°) and from a sample of squalene (m.p. 111.0–112.8°); mixture melting point 109.8–112.5°.

X-Ray diffraction data for squalene hexahydrochlorides. The values cited are for the *d*-spacing in Å and the corresponding intensity (I/I_1); squalene hexahydrochloride: 5.9 (0.40); 5.1 (0.70); 4.7 (1.00); 4.2 (0.18); 4.1 (0.13); 3.5 (0.07); 3.0 (0.09); hexahydrochloride of smoke component: 5.9 (0.38); 5.1 (0.70); 4.7 (1.00); 4.2 (0.20); 4.1 (0.16); 3.5 (0.08); 3.0 (0.18).

(12) A. I. Kosak, J. S. Swinehart, D. Taber, and B. L. Van Duuren, *Science*, **125**, 991 (1957).

(13) W. Carruthers and R. A. W. Johnston, *Chem. and Ind. (London)*, 1663 (1958).

(14) E. L. Wynder and G. F. Wright, *Cancer*, **10**, 255 (1957).

(15) L. F. Fieser and M. Fieser, *Steroids*, Reinhold Publishing Corp., New York, 1959, p. 352; E. R. H. Jones, P. A. Wilkinson, and R. N. Kerlogue, *J. Chem. Soc.*, 391 (1942); D. H. R. Barton and E. R. H. Jones, *J. Chem. Soc.*, 599 (1943).

(16) W. T. Beher, J. Parsons, and G. D. Baker, *Anal. Chem.*, **29**, 1147 (1957).

(17) W. T. Beher, J. Parsons, and G. D. Baker, *Henry Ford Hospital Med. Bull.*, **6**, 387 (1958).

(18) All melting points are corrected and were determined either on a Fisher-Johns block or a Nalge-Axelrod block. Rotations were measured in chloroform solution in a 1-dm. tube. Infrared spectra were determined in a Baird-Atomic Model 4-55 spectrophotometer with sodium chloride optics. X-ray diffraction data were obtained with a Norelco X-Ray Diffractometer with a copper target.

(19) A. C. Chibnall, S. H. Piper, H. A. Mangouri, E. F. Williams, and A. V. V. Iyengar, *Biochem. J.*, **31**, 1981 (1937).

(20) F. S. Kipping, *J. Chem. Soc.*, **57**, 980 (1890).

(21) H. J. Becker and J. Strating, *Rec. trav. chim.*, **59**, 933 (1940).

(22) The more usual Wolff-Kishner conditions left considerable amounts of unreduced carbonyl compound.

(23) I. Kondakov, *Bull. soc. chim. France*, **7**, 576 (1892).

(24) J. Hopper, *Analyst*, **72**, 513 (1947).

(25) Prepared as described in ref. 4.

Reduction of isolated squalene mixture. A solution of 3.5 mg. (0.0088 mmol.) of polyolefin in prereduced petroleum ether containing 5% rhodium on alumina catalyst absorbed 1.2 ml. of hydrogen (standard conditions).

Paper chromatography of squalene and related materials. Squalene, regenerated squalene,[10] the squalene mixture from smoke, and acid-treated squalene were spotted on Whatman No. 1 filter paper impregnated with "Quilon" (stearato chromic chloride[10,26]; the chromatogram was developed with methanol, and iodine vapor[27] was used for detection. R_f values were measured to the leading edge of the spot.

Isolation of sterol mixture from M. After the removal of polyolefins during silica gel chromatography of a 40 g. sample of M, the column (3.2 kg.) was eluted successively with 3 l. of benzene, 2.4 l. of 1:20 ethyl ether–benzene, 5 l. of 1:10 ethyl ether–benzene, 6 l. of 1:8 ethyl ether–benzene and 6 l. of 1:6 ethyl ether–benzene. The last solution yielded 0.21 g. of light orange oil which gave a positive Liebermann-Burchard test; crystallization from 1:1 benzene–methanol gave colorless crystals, m.p. 145.2–149.8°. The combined material from several runs was chromatographed on acid-washed alumina, and the column was eluted with petroleum ether, benzene, 4% ethyl ether in benzene, 12% ethyl ether in benzene, and 25% ethyl ether in benzene. The steroids were recovered from the last fraction; this procedure was repeated twice. The steroids were recrystallized from 4:1 methanol-ethanol and then from 4:1 petroleum ether–benzene to give colorless crystals, m.p. 158.4–162.8°. (In some runs it was found desirable to chromatograph the mixture once more on basic alumina at this point.) The yield of mixed steroids was of the order of 0.25% based on M.

Isolation and identification of β-sitosterol. This material was subjected to an extensive (more than 900 theoretical plates) fractional recrystallization from 1:1 benzene-methanol. Some of the filtrates from the previous recrystallizations were combined with various filtrates in the fractional recrystallization. From the filtrate side of the fractional recrystallization, sterol I was obtained, which after treatment with Darco and two recrystallizations from 95% ethanol gave rhombic platelets, m.p. 135–136°, $[\alpha]_D^{24}$ −38° (*c* 0.745). A mixture melting point of sterol I and β-sitosterol showed no depression. The acetate and benzoate of sterol I were made: acetate m.p. 127–128°, $[\alpha]_D^{25}$ −41.5° (*c* 0.823); benzoate m.p. 146–147°, $[\alpha]_D^{24}$ −14.7 (*c* 1.284); the reported constants for β-sitosterol are[28]: m.p. 137.0–137.5°, $[\alpha]_D^{24}$ −37°; for β-sitosteryl acetate: m.p.[29,30] 126.0–127.5°: $[\alpha]_D^{24}$ −42°[30]; for β-sitosteryl benzoate[28]: m.p. 146–147°, $[\alpha]_D^{24}$ −13.8°. The infrared spectra of the isolated sterol and its derivatives were identical with those of the corresponding sitosterol family.

Isolation and identification of stigmasterol. From the less soluble side of the fractional recrystallization, sterol II was obtained and recrystallized from ethanol to give colorless platelets, m.p. 169.5–170.5°, $[\alpha]_D^{24}$ −44.6°; a mixture melting point with an authentic sample of stigmasterol[31] was undepressed. The acetate of sterol II was prepared, m.p. 142.4–143.8°, $[\alpha]_D^{24}$ −53.6° (*c* 0.781), and the benzoate, m.p. 161.1–161.6°, $[\alpha]_D^{24}$ −24.2° (*c* 0.821). The reported constants for stigmasterol are: m.p. 170–171°,[32] $[\alpha]_D^{24}$ −45.8°[33]; for stigmasteryl acetate,[32] m.p. 144°, $[\alpha]_D^{20}$ −55.6°; for stigmasteryl benzoate,[34] m.p. 160.5–161.5°, $[\alpha]_D^{24}$ −24.5°.

Reduction of sterols. Sterols I and II and stigmasterol were separately reduced in acetic acid solution using a 5% rhodium on alumina catalyst (Baker and Co.). The moles of hydrogen taken up, respectively, per mole of steroid were 1.07, 2.14, and 2.10. The stanols and their acetates and benzoates were all identical as shown by melting points, mixture melting points, rotations, and infrared absorption spectra.

X-Ray diffraction data for sterols I and II. The values are in Å followed by intensity, I/I_1, in parentheses. Sterol I: 8.6 (0.10), 7.5 (0.10), 7.0 (0.13), 6.7 (0.16), 6.3 (0.12), 5.8 (0.75), 5.5 (1.00), 5.4 (0.87), 5.1 (0.37), 5.0 (0.24), 4.8 (0.26), 4.6 (0.18), 4.5 (0.17), 4.2 (0.19), 4.1 (0.14), 3.7 (0.12), 3.5 (0.12), 3.3 (0.08), 3.2 (0.07); for β-sitosterol: 8.6 (0.09), 7.4 (0.09), 7.0 (0.14), 6.7 (0.17), 6.3 (0.11), 5.8 (0.78), 5.5 (1.00), 5.4 (0.87), 5.1 (0.34), 5.0 (0.33), 4.8 (0.32), 4.6 (0.31), 4.5 (0.17), 4.2 (0.19), 4.1 (0.15), 3.7 (0.13), 3.5 (0.11), 3.3 (0.08), 3.2 (0.08); for sterol II: 7.4 (0.39), 6.7 (0.25), 6.2 (0.32), 5.9 (0.93), 5.1 (0.88), 4.8 (1.00), 4.6 (0.49), 4.3 (0.28), 4.0 (0.27), 3.9 (0.28), 3.6 (0.17), 3.5 (0.19), 2.7 (0.10), 2.3 (0.14), 2.3 (0.14), 2.2 (0.22); for stigmasterol: 7.4 (0.39), 6.7 (0.35), 6.2 (0.34), 5.9 (1.00), 5.1 (0.77), 4.8 (0.95), 4.6 (0.33), 4.3 (0.27), 4.0 (0.28), 3.9 (0.24), 3.6 (0.17), 3.5 (0.23), 2.7 (0.11), 2.4 (0.12), 2.3 (0.15), 2.2 (0.28).

Isolation of γ-sitosterol. The group of fractions closest to the four which yielded sterol I in the extended fractional recrystallization, yielded 0.47 g. of a sterol mixture which was acylated by refluxing with 32 ml. of acetic anhydride and 0.01 g. of sodium acetate. The mixed acetates were subjected to a 100-plate triangular fractional recrystallization from 1:2 benzene-ethanol. The six least soluble fractions were combined and yielded 3.4 mg. of a colorless compound, m.p. 142.3–143.6°, $[\alpha]_D^{23}$ −44.4° (*c* 0.31). This compound was hydrolyzed by refluxing with an aqueous ethanolic solution of potassium hydroxide under nitrogen to give sterol III which, after two recrystallizations from ethanol, melted at 145.0–146.4°, $[\alpha]_D^{22}$ −42.4° (*c* 0.014); the literature values for γ-sitosterol and its acetate are[35]: m.p. 147–148°, $[\alpha]_D$ −43°; m.p. 143–146°, $[\alpha]_D$ −46°.

Acknowledgment. We are greatly indebted to Dr. A. H. Boultbee and Mr. M. J. O'Neal of the Shell Oil Co. for the mass spectroscopic analyses. We also wish to thank Dr. S. Z. Lewin for his help with the x-ray diffractometer.

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(26) D. Kritchevsky and M. Calvin, *J. Am. Chem. Soc.*, **72**, 4330 (1950).

(27) D. Kritchevsky and M. R. Kirk, *Arch. Biochem. Biophys.*, **35**, 346 (1952).

(28) S. W. Gloyer and H. A. Schuette, *J. Am. Chem. Soc.*, **61**, 1901 (1939).

(29) J. W. Cook and M. Paige, *J. Chem. Soc.*, 337 (1944).

(30) E. S. Wallis and P. N. Chakravorty, *J. Org. Chem.*, **2**, 340 (1937).

(31) We are indebted to Dr. J. M. Chemerda of Merck and Co. for a generous gift of stigmasteryl acetate.

(32) R. Chakravorti and A. Dutta, *J. Indian Chem. Soc.*, **29**, 374 (1952).

(33) J. Simpson and N. E. Williams, *J. Chem. Soc.*, 737 (1937).

(34) A. C. Ott and C. D. Ball, *J. Am. Chem. Soc.*, **66**, 489 (1944).

(35) R. J. Anderson, R. L. Shriner, and G. O. Burr, *J. Am. Chem. Soc.*, **48**, 2992 (1926).