[CONTRIBUTION FROM THE NATIONAL INSTITUTE OF ARTHRITIS AND METABOLIC DISEASES, NATIONAL INSTITUTES OF HEALTH, PUBLIC HEALTH SERVICE, DEPARTMENT OF HEALTH, EDUCATION AND WELFARE]

# The Anthrasteroid Rearrangement. I. The Formation and Proof of Structure of Anthraergostapentaene[1]

BY WILLIAM R. NES AND ERICH MOSETTIG

RECEIVED FEBRUARY 15, 1954

The acid-catalyzed rearrangement at 23° of dehydroergosteryl acetate (Ib) to a *s*-octahydroanthracene derivative (II) is described. The structure of II has been elucidated by spectroscopic measurements and by dehydrogenation and oxidation experiments.

When dehydroergosteryl acetate (I) was treated in chloroform at 23° with hydrogen chloride in small concentrations (<0.07 *M*) a hydrocarbon II was obtained in yields of about 30%. The loss of the oxygen function was indicated by the infrared spectrum of II, by its behavior on the chromatographic column, and by the elemental analysis which agreed with the composition $C_{28}H_{40}$. The infrared spectrum indicated that the double bond in the side chain was still present and had not been involved in the rearrangement. The ultraviolet absorption revealed the presence of an aromatic ring with one conjugated double bond[2] (see Fig. 1). The aromatic ring was more clearly shown in the absorption spectrum of the tetrahydro derivative III. The

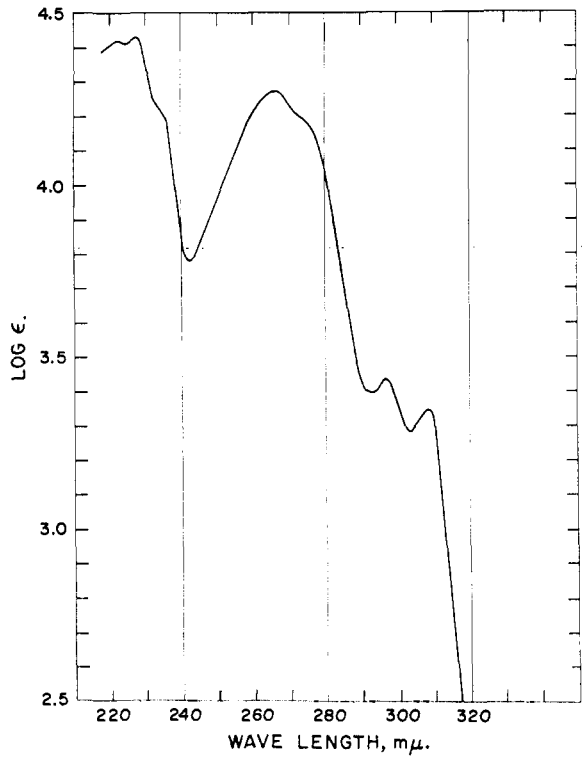
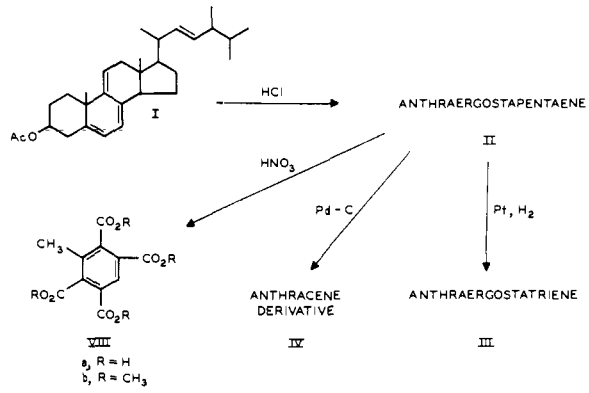



Fig. 1.—Absorption spectrum of anthraergostapentaene (II) in isoöctane.




three maxima (Fig. 2) at about 280 mμ with extinction coefficients less than 1000 can be attributed only to an aromatic system. The physical constants of II (m.p. 105–107°, $[\alpha]_D$ −70°) eliminated the possibility that it was the aromatic "neoergostapentaene" described by Cook and Haslewood[3] (m.p. 93–94°, $[\alpha]_D$ +51°), and by Windaus and Deppe[4] (m.p. 89–90°, $[\alpha]_D$ +68°). The large difference in rotation between II and "neoergostapentaene" also suggested that II was not merely the methyl homolog of "neoergostapentaene." Furthermore, hydrocarbon II showed a large positive rotational difference for the saturation of the two non-aromatic double bonds, while the analogous saturation of "neoergostapentaene" gives a large negative difference.[5] Similar considerations made a structure analogous to the "$\Delta^{11}$-neoergosterol" of Windaus and Roosen-Runge[2b] appear unlikely. We have now established that II is not a steroid at all, but is a derivative[6] of *s*-octahydroanthracene with one double bond in conjugation with the aromatic ring. This was shown by ultraviolet spectrophotometry and by dehydrogenation and oxidation experiments.

An analysis of the absorption of several model compounds revealed that the maxima of highest intensity for *s*-octahydroanthracene and for each of its derivatives shown in Figs. 2 and 3 are at wave lengths above 280 mμ with extinction coefficients greater than 570, while those for *s*-octahydrophen-

(1) A preliminary report of this work has been published in THIS JOURNAL, **75**, 2787 (1953). Presented in part at the 124th National Meeting of the American Chemical Society, September 6–11, 1953.

(2) *Cf.* the absorptions of (a) "neoergostapentaene" and of its 22-dihydro derivative (G. A. D. Haslewood and E. Roe, *J. Chem. Soc.*, 465 (1935)), (b) $\Delta^{11}$-neoergosterol (A. Windaus and C. Roosen-Runge, *Ber.*, **73**, 321 (1940)) and (c) 1,2-dihydronaphthalene (R. A. Morton and A. J. A. de Gouveia, *J. Chem. Soc.*, 916 (1934)).

(3) J. W. Cook and G. A. D. Haslewood, *Chemistry and Industry*, **53**, 507 (1934).

(4) A. Windaus and M. Deppe, *Ber.*, **70**, 76 (1937).

(5) See K. Bonstedt, *Z. physiol. Chem.*, **185**, 165 (1929), for the rotation of the tetrahydro derivative.

(6) We propose that the perhydro compound bearing no side chain be called anthrastane, the compound with the ergosterol side chain anthraergostane, and that with the cholesterol side chain anthracholestane, etc., since features of both an anthracene and a steroid are present. Accordingly, we shall call the conversion of a steroid to such a product the "anthrasteroid rearrangement."

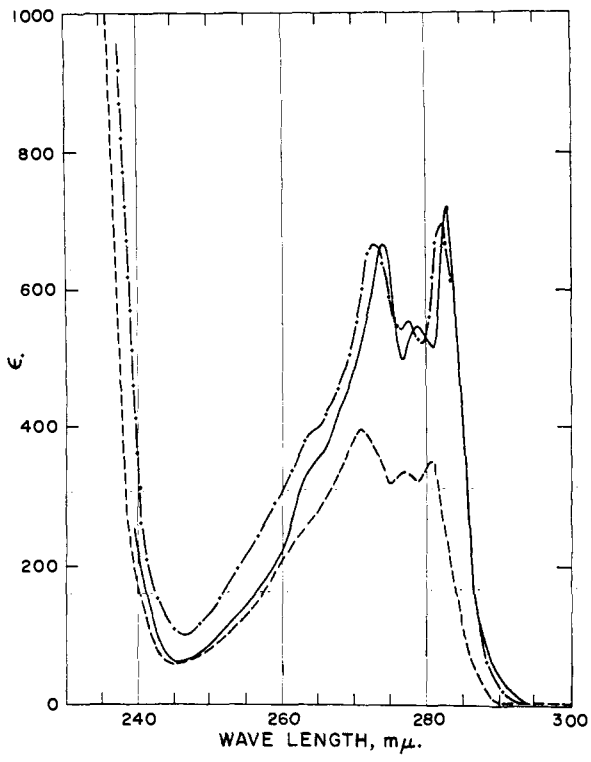



Fig. 2.—Absorption spectra in isoöctane: – – – –, 9-methyl-*s*-octahydrophenanthrene[9]; ———, 9-methyl-*s*-octahydroanthracene[9]; – · – · –, anthraergostatriene (III).

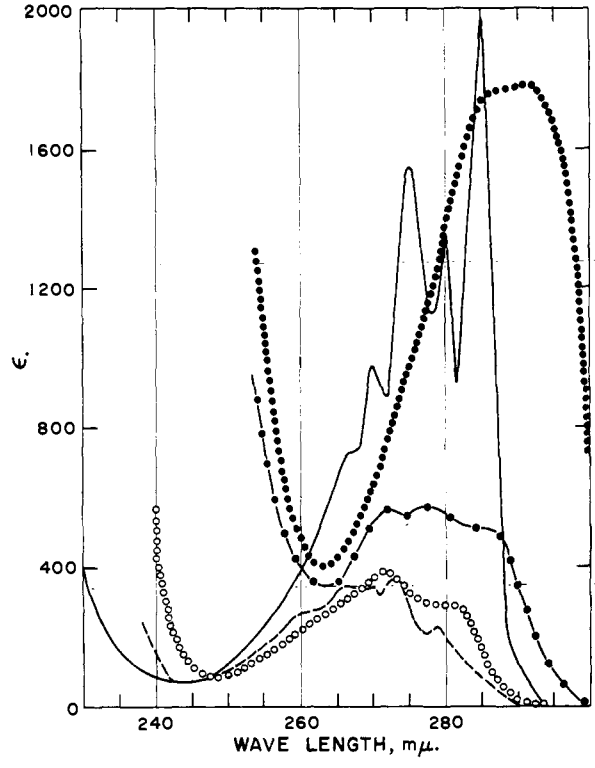



Fig. 3.—Absorption spectra in isoöctane: – – – –, *s*-octahydrophenanthrene (prepared according to J. R. Durland and H. Adkins, THIS JOURNAL, **59**, 135 (1937)); –●–●–●–, 9-chloromethyl-*s*-octahydrophenanthrene[9]; O O O O, 9-hydroxymethyl-*s*-octahydrophenanthrene[9]; ———, *s*-octahydroanthracene[9]; ● ● ● ●, 9-chloromethyl-*s*-octahydroanthracene.[9]

anthrene and each of its derivatives (Figs. 2 and 3) are below 280 mμ with extinction coefficients smaller than 570. The maximum of highest intensity (693) of anthraergostatriene (III) is at 282 mμ and, therefore, should be derived from *s*-octahydroanthracene. Figure 2 demonstrates clearly the similarity of the spectrum of III with the spectrum of 9-methyl-*s*-octahydroanthracene and its dissimilarity with the spectrum of 9-methyl-*s*-octahydrophenanthrene.

When we submitted either II or III to dehydrogenation with palladium–charcoal at temperatures not exceeding 300°, a compound was formed that showed an ultraviolet spectrum typical of a fully aromatic anthracene derivative IV. The elemental composition of IV ($C_{27}H_{34}$) agreed with that of a methylcyclopentanthracene carrying a saturated ergosterol side chain. In addition another compound V was isolated which probably contained more aromatic nuclei. In order to show that a rearrangement to an anthracene derivative could not have taken place during the dehydrogenation, we subjected a steroid containing an aromatic B-ring (neoergosterol) to the same dehydrogenating conditions. In this instance we isolated only a phenanthrene derivative VI and a naphthofluorene derivative VII.[7] The elemental composition of VI ($C_{26}H_{32}$) agreed with that of a cyclopentanophenanthrene having a saturated ergosterol side chain. The absorption spectrum was identical with that of 1,2-cyclopentanophenanthrene in the region from 220 to 400 mμ. The elemental composition, melting point and spectrum of VII were identical with those of 5-[2-(3-methylbutyl)]-7-methyl-2′,1′-naphtho-1,2-fluorene.[8]

When the anthraergostapentaene II was refluxed in concentrated nitric acid overnight, 1-methyl-2,3,5,6-tetracarboxybenzene (VIII)[9] was produced. This acid could have arisen only from an anthracene-like structure. The possibility of a rearrangement during the oxidation can be excluded since Inhoffen[10] has shown that neoergosterol yields 1,2,3,4-tetracarboxybenzene. The presence of the methyl group on the aromatic nucleus of II and III was further proved by oxidation of II in dilute nitric acid at 195° to pentacarboxybenzene (IX).

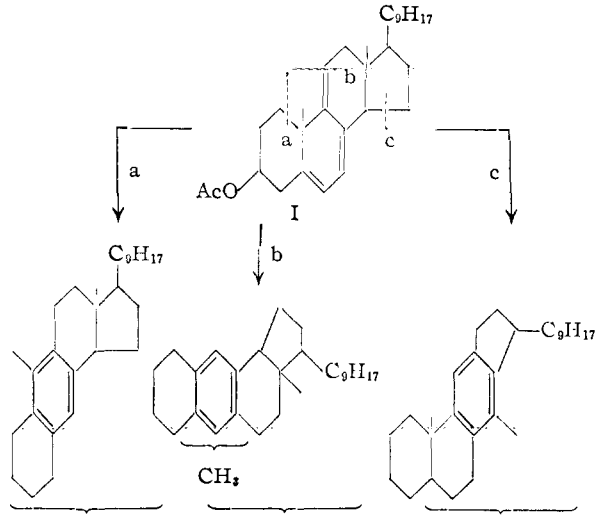
A hydrocarbon having the 9-methyl-*s*-octahydroanthracene skeleton could arise from dehydroergosteryl acetate (I) only by two types of carbon–carbon rupture, namely, at $C_1$–$C_{10}$ (a) and $C_9$–$C_{11}$ (b), leading to formulations A and B, respectively, for hydrocarbon II. A break at $C_{14}$–$C_{15}$ (c) could give the structure C which also might satisfy the spectral data and oxidation results.

(7) H. Honigmann, *Ann.*, **511**, 292 (1934), has dehydrogenated neoergosterol under similar conditions but isolated only the phenolic fraction of the reaction mixture. In our investigation we made no attempt to obtain phenolic material.

(8) (a) L. F. Fieser and M. Fieser, "Natural Products Related to Phenanthrene," 3rd ed., Reinhold Publ. Corp., New York, N. Y., 1949, pp. 155–156; (b) W. V. Mayneord and E. M. F. Roe, *Proc. Roy. Soc. (London)*, **A152**, 299 (1935).

(9) W. R. Nes and E. Mosettig, THIS JOURNAL, **76**, 3186 (1954).

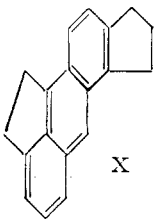
(10) H. H. Inhoffen, *Ann.*, **497**, 130 (1932).




It is unlikely, however, that dehydrogenation of C would produce an anthracene derivative, and, if so, a benzanthracene might have been expected. Furthermore, the rearrangement in the 10-epi series[11] led to the same compound as in the series having the natural configuration at $C_{10}$; had the rearrangement proceeded according to scheme (c) a diastereoisomer should have been obtained. A choice between formulations A and B must await further evidence. However, it is interesting that the rearrangement in the 10-epi series did not produce an aromatic B-ring *steroid* by methyl ($C_{19}$) migration to positions 6 or 7. In agreement with these findings is the fact that lumisterol yields 1-methyl-2,3,-5,6-tetracarboxybenzene[9,12] and not the isomeric compound on oxidation with nitric acid. If further evidence proves that formulation A is correct, this would indicate that the reaction is not stereospecific at $C_{10}$.

It was interesting to ascertain whether ergosterol itself is rearranged to an anthrasteroid as well as to the ergosterol-B isomers. We have treated ergosterol with HCl in chloroform and have failed to detect any hydrocarbon such as the dihydro derivative of II. That dehydration to a (steroidal) hydrocarbon is actually the first step in the anthrasteroid rearrangement will be shown in a forthcoming communication. Inhoffen[13] has also raised some questions relative to the scope and mechanism of this rearrangement which suggest further investigations along these lines.

Since many of the potent carcinogens are anthracene derivatives with a ring attached at the 1,2-positions (*e.g.*, 10-methyl-1,2-benzanthracene), it is not unreasonable to speculate that this type of facile transformation, *i.e.*, steroids into hydroanthracene derivatives, forms part of a biogenetic route to endogenous carcinogens. A structural resemblance to compounds II, III and IV is found in 1,2-cyclopentano-5,10-aceanthracene (X) which shows moderate but definite carcinogenic activity.[14]




Compounds II, III and IV and their analogs are being examined for carcinogenic activity in mice by Dr. Murray J. Shear of the National Cancer Institute.

## Experimental[15]

Anthraergostapentaene (II).—To a solution of 6.00 g. of dehydroergosteryl acetate (I) in 250 ml. of chloroform was added 50.0 ml. of 0.3 *N* hydrogen chloride in chloroform at 23°. When the value of the extinction coefficient at 266 mμ had become essentially constant (about 80 min.), the dark green reaction mixture was extracted with aqueous sodium bicarbonate. The color changed to red when all of the HCl had been extracted. The solution was then washed with water, dried over sodium sulfate and the solvent was removed in a vacuum. The residue was dissolved in 250 ml. of light petroleum ether and adsorbed on a column of 110 g. of aluminum oxide. Elution with 500 ml. of 5% benzene in light petroleum ether yielded, after evaporation of the solvent, 1.8 g. of material. This was recrystallized from ethyl acetate–ethanol to give 1.57 g. (30%) of slightly yellow rosettes, m.p. 100–102°. Further recrystallization from acetone failed to remove the color completely, but raised the melting point to 105–107°, $[\alpha]D$ −70°, $\lambda_{max}$ 222, 227, 266, 296 and 308 mμ ($\epsilon$ 26,100, 27,100, 18,600, 2,760 and 2,220, respectively) (see Fig. 1), $\lambda_{max}$ 968 cm.$^{-1}$.

*Anal.* Calcd. for $C_{28}H_{40}$: C, 89.29; H, 10.70. Found: C, 88.96; H, 10.74.

Stability of Anthraergostapentaene (II) to Rearrangement Conditions.—A solution of II in chloroform, which was 0.05 *M* in hydrocarbon, 0.04 *M* in HCl and 0.05 *M* in acetic acid, was allowed to stand at 20° two hours. Aliquots were withdrawn at intervals, evaporated to dryness and the ultraviolet spectrum determined in isoöctane. No change was observed.

This experiment shows that the low yield of II is caused by the formation of by-products and not by the decomposition, polymerization, etc., of II after it is formed.

Anthraergostatriene (III).—A solution of 1.00 g. of II in 35 ml. of ethyl acetate and 16 ml. of glacial acetic acid was shaken in a hydrogen atmosphere with 198 mg. of platinum oxide. Two molar equivalents of hydrogen was absorbed in ten minutes after which the rate of absorption was practically zero. The solvents were removed in a vacuum. The residue was crystallized from *ca.* 20 ml. of acetone by refrigeration at −20°. The product weighed 0.84 g. (82%)

(11) Starting with lumisteryl acetate, the 10-epi-$\Delta^{5,7,9(11)}$-cholestatrienyl acetate was obtained only as a mixture with what may be the starting material. It is remarkable that, contrary to the usual course of the mercuric acetate dehydrogenation at $C_9$–$C_{11}$ in which not more than two moles of mercurous acetate is produced (W. V. Ruyle, T. A. Jacob, J. M. Chemerda, E. M. Chamberlin, D. W. Rosenburg, G. E. Sita, R. L. Erickson, L. M. Aliminosa and M. Tishler, THIS JOURNAL, **75**, 2604 (1953), and unpublished observations from this Laboratory), the reaction with lumisteryl acetate readily produced as much as five moles of mercurous acetate. A crystalline product could be obtained only when less than about three moles precipitated. This product showed absorption maxima at *ca.* 280 and 320 mμ indicating a $\Delta^{5,7}$-diene system and a $\Delta^{5,7,9(11)}$-triene system.

(12) H. H. Inhoffen, *Ann.*, **494**, 124 (1932).

(13) H. H. Inhoffen, *Naturwissenschaften*, **40**, 455 (1953).

(14) M. J. Shear and J. Leiter, *J. Natl. Cancer Inst.*, **2**, 99 (1941).

(15) All melting points were determined on a Kofler block and are recorded as read. Rotations were determined at 20° in chloroform in *ca.* 1% solutions and are accurate to within about 2°. All chloroform used in this work, including that for reactions, rotations and chromatography, contained 0.75% ethanol (Merck, Reagent). Ultraviolet spectra were determined in "spectro grade" isoöctane on a Cary recording spectrophotometer. Infrared spectra were determined in carbon disulfide (unless otherwise noted) on a Perkin–Elmer double beam spectrophotometer by Mrs. Alma L. Hayden and Mrs. Phyllis B. Smeltzer. Analyses are by the Analytical Service Laboratory of this Institute under the direction of Dr. William C. Alford.

and formed in colorless, transparent, rectangular plates, m.p. 105–107°. After several recrystallizations the melting point was 106–107°, $[\alpha]_D$ +21°, $\lambda_{max}$ 273, 278 and 282 mμ (ε 670, 550 and 695, respectively), $\lambda_{infl.}$ 264 mμ (ε 387), $\lambda_{min}$ 247 mμ (ε 105).

*Anal.* Calcd. for $C_{28}H_{44}$: C, 88.34; H, 11.65. Found: C, 88.42; H, 11.47.

**Dehydrogenation of II and III.**—A mixture of 0.40 g. of II and 0.20 g. of 5% palladium-on-charcoal was heated for 2.5 hours at 220–300° and then for 2.5 hours at 300° during which time 87 ml. (standard conditions) of gas was evolved. The cooled reaction mixture was extracted with ether, and the solvent was removed. Chromatography of the residue on 18 g. of $Al_2O_3$ yielded an oil by elution with 600 ml. of 5% benzene in light petroleum ether. Rechromatography gave a nearly colorless oily **anthracene derivative (IV)** which could not be crystallized, $\lambda_{max}$ 227, 235, 262.5, 338, 354, 372 and 391 mμ (ε 20,100, 21,100, 133,000, 2,540, 4,230, 5,910, 5,910, respectively). The yield of IV was about 25%. Material with the same spectrum was obtained by a similar dehydrogenation of III.

*Anal.* Calcd. for $C_{27}H_{34}$: C, 90.44; H, 9.55. Found: C, 90.53; H, 9.48.

From the original chromatogram another compound V was obtained by elution with benzene. It was also easily obtained in small amounts by treatment of a dehydrogenation mixture with 2,4,7-trinitrofluorenone and subsequent decomposition of the complex on alumina. From acetone V forms as pale yellow crystals, m.p. 179–182°, $\lambda_{max}$ 234, 270, 290, 296, 302, 312, 342, 358, 368, 376 and 386 mμ (for 0.167 mg. in 50.0 ml. of isoöctane, 2.0 cm. cells, D 0.59, 0.85, 1.10, 1.18, 1.31, 0.72, *ca.* 0.06, 0.10, 0.10, 0.10, 0.10, respectively). Because of the small amounts available we were unable to purify the compound completely. Compound V was also apparent spectroscopically in the mixture resulting from the dehydrogenation of anthraergostatriene (III). Extensive analysis of various dehydrogenation mixtures failed to show the presence of a phenanthrene derivative. The anthracene derivative IV was usually present in about a 25% yield.

**Dehydrogenation of Neoergosterol.**—A mixture of 1.0 g. of neoergosterol and 0.5 g. of 5% palladium-on-charcoal was heated for 1.5 hours at 220–240° and then at 300° for an additional six hours. Approximately 4 moles of gas (240 ml. at standard conditions) per mole of steroid was evolved. The mixture was cooled and extracted with ether. Evaporation of the solvent left an oil which was dissolved in 200 ml. of light petroleum ether and adsorbed on 30 g. of aluminum oxide. Elution was as follows: (1) 200 ml. of light petroleum ether; (2) 200 ml. of 2% benzene in light petroleum ether; (3) 600 ml. of 5% benzene in light petroleum ether; (4) 250 ml. of 10% benzene in light petroleum ether; (5) 200 ml. of 100% benzene.

Fraction 3 was evaporated to dryness. The resulting oil was crystallized from acetone–methanol three times to give the phenanthrene derivative VI as clusters of colorless crystals melting unsharply at 105°; $\lambda_{max}$ 225, 260, 281, 288, 301, 320, 327, 335, 342 and 350 mμ (ε 28,000, 67,000, 16,200, 12,400, 15,000, 900, 500, 1,100, 400 and 1,200, respectively).

*Anal.* Calcd. for $C_{26}H_{32}$: C, 90.63; H, 9.36. Found: C, 90.49; H, 9.52.

In another similar experiment a more extensive chromatogram of the dehydrogenation mixture was made and an anthracene derivative could not be detected in any fraction. According to spectroscopic measurements the yield of VI in the fraction corresponding to fraction 3 above was 15–20%.

Evaporation of fraction 5 yielded a substance which was crystallized from acetone. Recrystallization yielded VII as nearly colorless needles, m.p. 210–212°, $\lambda_{max}$ 232, 239, 272, 282, 299, 307, 314, 320, 333, 341, 349, 357 and 365 mμ (ε 27,600, 27,000, 58,600, 74,500, 37,900, 29,600, 22,000, 35,200, 2,100, 1,100, 2,300, 900 and 2,500, respectively).

The melting point reported for 5-[2-(3-methylbutyl)]-7-methyl-2′,1′-naphtho-1,2-fluorene is 214–215°.[8a]

*Anal.* Calcd. for $C_{27}H_{26}$: C, 92.00; H, 8.00. Found: C, 92.36; H, 7.56.

**1-Methyl-2,3,5,6-tetracarbomethoxybenzene (VIIIb from II).**—The hydrocarbon II (1.00 g.) was refluxed overnight with 40 ml. of 70% nitric acid. Removal of 30 ml. by distillation caused precipitation. The mixture was refrigerated and then filtered to yield 126 mg. of crude acid. Esterification with diazomethane and recrystallization of the product from methanol–water yielded needles of 1-methyl-2,3,5,6-tetracarbomethoxybenzene, m.p. 121–123°. The mixture with an authentic sample[9] melted at the same temperature and the infrared spectra of the two samples were identical.

*Anal.* Calcd. for $C_{15}H_{16}O_8$: C, 55.55; H, 4.97. Found: C, 55.43; H, 5.06.

**Pentacarbomethoxybenzene (IX from II).**—One-half gram of the hydrocarbon II was heated with 10.0 ml. of water and 5.0 ml. of nitric acid (70%) in a sealed tube at 195° for 20 hours. The pale yellow solution was evaporated to dryness in a vacuum and the residue was esterified with diazomethane. The resulting ester was recrystallized from methanol and methanol–water to give needles, m.p. 147–148°. The infrared spectrum (Nujol) was identical with that of an authentic sample. The reference spectrum was obtained from Samuel P. Sadtler and Sons.

*Anal.* Calcd. for $C_{16}H_{16}O_{10}$: C, 52.17; H, 4.37. Found: C, 52.14; H, 4.58.

**Mercuric Acetate Dehydrogenation of 10-Epi-$\Delta^{5,7}$-cholestadienyl Acetate (Lumisteryl$_3$ Acetate).**—A solution of 6.0 g. (0.014 mole) of the diene acetate[16] in 30 ml. of carbon tetrachloride and 100 ml. of ethanol was refluxed two hours with 14 g. (0.044 mole) of mercuric acetate in 14 ml. of acetic acid and 40 ml. of ethanol. The solution became pink and finally yellow and mercurous acetate began to precipitate almost immediately. The mixture was filtered to yield 8.4 g. (0.033 mole) of mercurous acetate. The filtrate was diluted with water and extracted with carbon tetrachloride. After several recrystallizations from acetone–methanol the product (0.65 g.) formed as colorless, flaky crystals, m.p. 129–131°. The compound showed absorption maxima (mμ) at 270, 280, 320 and an inflection at 338 mμ, and the relative intensities indicated *ca.* 20% $\Delta^{5,7,9(11)}$-triene and *ca.* 80% $\Delta^{5,7}$-diene.

*Anal.* Calcd. for $C_{29}H_{44}O_2$: C, 82.01; H, 10.45. Found: C, 81.89; H, 10.69.

The above procedure is used routinely in this Laboratory to give relatively good yields of dehydroergosteryl acetate from ergosteryl acetate. The diene–triene mixture, on treatment with HCl in chloroform, yielded crystals, m.p. 123–125°, in over 50% yield. An acetoxyl group was evident from the infrared spectrum and a heteroannular diene system from the ultraviolet spectrum ($\lambda_{max}$ 249 mμ). These data indicate the product was one or more of the B isomers of the original starting material. The ultraviolet spectrum also indicated ($\lambda_{max}$ 296 and 308 mμ) a small amount of anthracholestatetraene (see below). In further attempts to obtain the $\Delta^{5,7,9(11)}$-triene pure, many experiments were run. On the supposition that an intermediate diacetate product might yield the desired compound by pyrolysis,[17] we submitted a reaction mixture to a temperature of 150–190° for ten minutes[17] without any effect on the ultraviolet spectrum. Pyrolysis at 180° for one hour changed the spectrum, but the desired compound could not be obtained by chromatography. When the mercuric acetate dehydrogenation was carried out at room temperature in acetic acid–chloroform for 16 hours, the mercurous acetate (analyzed for mercury in one case) which precipitated corresponded to two to five molecular equivalents (based on the steroid) depending on the ratio of mercuric acetate used to start with.

**Anthracholestatetraene.**—A solution of 0.52 g. of impure 10-epi-$\Delta^{5,7,9(11)}$-cholestatrienyl acetate (assay (ultraviolet): *ca.* 20% $\Delta^{5,7,9(11)}$, 80% $\Delta^{5,7}$) in 14 ml. of chloroform was allowed to stand with 7 ml. of *ca.* 0.3 *M* HCl in chloroform for 30–60 minutes at room temperature. After the solvent was removed in vacuum, the residue was adsorbed on 15 g. of alumina. Elution with 100 ml. of 5% benzene in light petroleum ether gave an oil which readily crystallized from acetone–methanol. The anthracholestatetraene so obtained weighed 17 mg. and formed in nearly colorless rosettes, m.p. 114–115°, $[\alpha]_D$ −35°, $\lambda_{max}$ 222, 227, 266, 296 and 308 mμ (ε 23,500, 24,800, 15,900, 2,380 and 1,860). This material did not depress the melting point of an authentic

(16) The 3,5-dinitrobenzoate was obtained from the Sterling–Winthrop Research Institute through the courtesy of Dr. C. M. Suter. The acetate was prepared from this by hydrolysis and acetylation.

(17) See reference in footnote 11.

sample[18] with the same physical constants, and the infrared spectra were identical.

*Anal.* Calcd. for $C_{27}H_{40}$: C, 88.94; H, 11.06. Found: C, 89.16; H, 11.20.

**Treatment of Ergosteryl Acetate with HCl.**—A solution (27 ml.) of ergosteryl acetate (6.1 g.) in chloroform, which was 0.06 *M* in HCl, was allowed to stand at room temperature. The ultraviolet spectrum was unchanged after four hours and 25 ml. of 0.47 *M* HCl was added. The spectrum of the $\Delta^{5,7}$-diene system had completely disappeared after six hours and was replaced by the spectrum ($\lambda_{max}$ 250 m$\mu$) of the B-isomers. The dark green solution was evaporated to dryness and the residue was crystallized from ether–methanol to yield 4.5 g. of the B-isomers as colorless needles melting unsharply at 100°, $\lambda_{max}$ 250 m$\mu$ ($\epsilon$ 20,000). Fieser, *et al.*,[19] report $\lambda_{max}$ 250 m$\mu$ ($\epsilon$ *ca.* 20,000) for ergosteryl-$B_1$ acetate. The mother liquor was chromatographed on alumina, but only traces of material were eluted by 5 and 10% benzene in petroleum ether which should have removed a hydrocarbon. An additional amount of the B-isomers was eluted by benzene.

(18) A forthcoming publication from this Laboratory will describe the preparation of anthracholestatetraene starting with 7-dehydrocholesterol.

(19) M. Fieser, W. E. Rosen and L. F. Fieser, THIS JOURNAL, **74**, 5397 (1952).

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[CONTRIBUTION FROM THE NATIONAL INSTITUTE OF ARTHRITIS AND METABOLIC DISEASES, NATIONAL INSTITUTES OF HEALTH, PUBLIC HEALTH SERVICE, DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE]

# The Anthrasteroid Rearrangement. II. The Structural Proof of 1-Methyl-2,3,5,6-tetracarboxybenzene[1]

BY WILLIAM R. NES AND ERICH MOSETTIG

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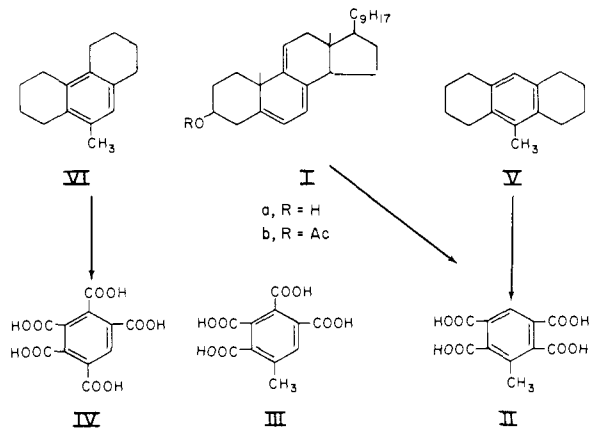
The acid which is obtained by the nitric acid oxidation of steroids containing two double bonds in ring B was found to be identical with the acid obtained from 9-methyl-*s*-octahydroanthracene. The acid is, therefore, 1-methyl-2,3,5,6-tetracarboxybenzene. The oxidation of 9-methyl-*s*-octahydrophenanthrene yielded pentacarboxybenzene. 9-Methyl-*s*-octahydrophenanthrene has been characterized by several new physical constants and has been prepared by a new route involving the previously unreported 9-hydroxymethyl-*s*-octahydrophenanthrene.

The structural proof of anthraergostapentaene[2] depends in part on the assignment of a correct formula to the methyltetracarboxybenzene of Inhoffen[3] and others.[4–6] In this paper the unequivocal preparation of this acid is described.

Although the use of nitric acid for degrading polynuclear hydrocarbons to an aromatic acid is usually applied to compounds containing one or more aromatic rings,[7] this technique has been successfully employed with non-aromatic compounds.[3–6] Thus it has been found that steroids containing two double bonds in ring B, *e.g.*, ergosterol, $\Delta^{6,8}$-coprostadienol and dehydroergosterol (Ia), yield an aromatic acid, while steroids in which ring B is not intact do not give this material. The methyl ester of this acid (prepared with diazomethane[3,6]) melts at 123–124°. Inhoffen[3] has shown that this acid must be either 1-methyl-2,3,5,6-tetracarboxybenzene (II) or 1-methyl-2,3,4,5-tetracarboxybenzene (III), since it forms a dianhydride and could be oxidized to pentacarboxybenzene (IV). For reasons which are not stated, later papers on the subject[5,6] ignore structure II and refer to this acid as III. Müller[6] actually misquotes Inhoffen on this subject. Feist[8] has reported the synthesis of II, but his methyl ester (m.p. 103–104°) was prepared with methanolic HCl which usually does not lead to a neutral ester,[9] and he gives no elemental analysis.




By oxidation of the appropriate *s*-octahydroanthracene and *s*-octahydrophenanthrene derivatives acids II and III, respectively, should be obtained. We have prepared 9-methyl-*s*-octahydroanthracene (V), essentially according to the directions of Badger, *et al.*,[10] and submitted it to oxidation with nitric acid. In order to obtain the reference acid, we oxidized dehydroergosteryl acetate (Ib). The products from the two oxidations were identical, and Inhoffen's acid is, therefore, 1-methyl-2,3,5,6-tetracarboxybenzene (II). It was hoped that in the analogous oxidation of 9-methyl-*s*-octahydrophenanthrene (VI) the methyl group would be unaffected and that the isomeric acid III would result. However, the only identifiable polycarboxylic acid was pentacarboxybenzene (IV). While both V and II have two *ortho* substituents, VI and III have only one substituent *ortho* to the methyl group, and this difference in steric hindrance may be

(1) A preliminary report of this work has been published in THIS JOURNAL, **75**, 2787 (1953). Presented in part at the National Meeting of the American Chemical Society, September 6–11, 1953.

(2) W. R. Nes and E. Mosettig, *ibid.*, **76**, 3182 (1954).

(3) H. H. Inhoffen, *Ann.*, **494**, 122 (1932).

(4) F. Reindel and K. Niederländer, *ibid.*, **482**, 264 (1930).

(5) A. Windaus and G. Zühlsdorff, *ibid.*, **536**, 204 (1938).

(6) M. Müller, *Z. physiol. Chem.*, **233**, 223 (1935).

(7) *Cf.* W. P. Campbell, M. D. Soffer and T. R. Steadman, THIS JOURNAL, **64**, 425 (1942).

(8) F. Feist, *Ann.*, **496**, 99 (1932).

(9) D. E. Read and C. B. Purves, THIS JOURNAL, **74**, 116 (1952).

(10) G. M. Badger, W. Carruthers, J. W. Cook and R. Schoental, *J. Chem. Soc.*, 169 (1949).