the fractions obtained by ozonolysis of parthenin at room temperature, which also results in the formation of formic acid, gave a positive iodoform test and the n.m.r. spectrum of parthenin had no signal corresponding to the $=C-CH_3$ peaks present, for example, in the n.m.r. spectra of dihydroparthenin (II), anhydroparthenin (VI), neotenulin and santonin.

One of the several reduction products of VI, hexahydroanhydroparthenin (VII), proved to be identical with tetrahydroambrosin.[7,8] This provides proof for the previously assumed structure of ambrosin (VIII)[7] and shows that I and VIII have the same stereochemistry at C_4, C_5, C_6 and C_7. The rotatory dispersion curve[9] of III is almost superimposable on that of tetrahydrohelenalin which suggests the absolute configuration at C_1, C_4 and C_5.

Acknowledgment.—This investigation was supported by a grant from the United States Public Health Service (RG-5814).

(7) (a) H. Abu-Shady and T. D Soine, *J. Am. Pharm. Assoc.*, **42**, 387 (1953); **43**, 365 (1954); (b) L. Bernardi and G. Büchi, *Experientia*, **13**, 466 (1957); (c) F. Šorm, M. Suchý and V. Herout, *Coll. Czechoslov. Chem Commun.*, **24**, 1548 (1959).

(8) We wish to thank Dr. V. Herout for carrying out the comparison.

(9) Kindly determined by Professor C. Djerassi.

(10) Recipient of a Fulbright Travel Grant 1958–1959.

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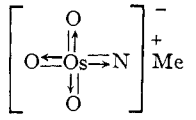
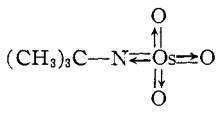
ORGANIC OSMIAMATES[1]

Sir:

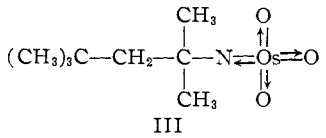
Osmiamates (I)[2] in which the osmium is octovalent are not known in Organic Chemistry. We wish to report the synthesis of two organic osmiamates: *t*-butyl osmiamate (II) and 1,1,3,3-tetramethylbutyl osmiamate (III) which were ob-



I



II



III

tained as a result of our general studies on the reaction of osmium tetroxide with various groups of organic compounds.[3,4] *t*-Butyl osmiamate was prepared by allowing osmium tetroxide (1.0 g.) in 50 cc. of pure ligroin to drop slowly with stirring at 0° and preferably in a nitrogen atmosphere into a ligroin solution (25 cc.) of excess (7.0 g.) of *t*-butylamine. Stirring was continued for 24 hours whereby an orange precipitate separated out and was removed by filtration. This was recrystallized several times from hot pure *n*-pentane at −10° into hair-like, orange-yellow crystals which agglomerated like cotton fibers; yield, 65%; m.p. 110°. This compound also can be prepared in aqueous solutions.

Anal. Calcd. for $C_4H_9NO_3Os$: N, 4.53; Os, 61.48; mol. wt., 309. Found: N, 4.68; Os,[5,6] 60.08; mol. wt., 303 (cryoscopic in benzene).

With the thiourea reagent[3] *t*-butyl osmiamate gave an immediate pink coloration characteristic for octovalent osmium. A paper chromatogram developed with ligroin–*t*-butyl alcohol mixture, 90:10 v./v., gave an R_f of 0.68 (32°). An infrared spectrum 10% in $CHCl_3$ showed a strong band, absent in the infrared spectrum of the amine, at 910–915 cm.$^{-1}$ compared with that of osmium tetroxide at 951 cm.$^{-1}$. Bands usually attributed to the amino or hydroxyl hydrogens were absent. An ultraviolet spectrum in *t*-butyl alcohol gave a maximum at 323 mμ; ϵ, 4066.

1,1,3,3-Tetramethylbutyl osmiamate (III) was prepared in exactly the same way as the *t*-butyl osmiamate; yield, 69%; m.p. 51.5° (*n*-pentane).

Anal. Calcd. for $C_8H_{17}NO_3Os$: N, 3.83; Os, 52.04. Found: N, 3.80; Os, 51.89.

This osmiamate also gave an immediate pink color with the thiourea reagent and on a paper chromatogram an R_f value of 0.87 (32°). The infrared spectrum 10% in $CHCl_3$ showed a strong band at 910–915 cm.$^{-1}$ with the amino and hydroxyl hydrogen bands absent. The ultraviolet spectrum in *t*-butyl alcohol showed a strong band with a maximum at 323 mμ; ϵ, 3650.

Both osmiamates react with dilute sulfuric acid to give osmium tetroxide and the sulfates of the original amines. They deflagrate spontaneously on a hot plate and show strong oxidizing properties; they react with olefins in the same manner as osmium tetroxide. These and other reactions of osmiamates are now being investigated and will be reported later.

Acknowledgment.—The authors are indebted to Dr. Shin-ichi Sasaki and Mrs. J. A. Hilton for technical assistance, to Dr. S. Nagy for the nitrogen analysis and to Rohm and Haas for a supply of 1,1,3,3-tetramethylbutylamine.

(1) Supported by NIH Contract B-1493(C1).

(2) N. V. Sidgwick, "The Chemical Elements and Their Compounds," Vol. II, Oxford University Press, 1950, p. 1507.

(3) N. A. Milas, J. H. Trepagnier, J. T. Nolan, Jr., and M. I. Iliopulos, THIS JOURNAL, **81**, 4730 (1959).

(4) N. A. Milas and M. I. Iliopulos, NIH Report, March (1959).

(5) W. Geilmann and R. Neeb, *Z. anal. Chem.*, **156**, 420 (1957).

(6) R. Criegee, *Ann.*, **522**, 75 (1936); R. Criegee, B. Marchand and H. Wannowius, *ibid.*, **550**, 99 (1942). Criegee, *et al.*, indicated that osmium analyses gave always low values.

(7) NIH Postdoctorate Research Associate, Fulbright Traveling Fellow.

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THE BIOLOGICAL CONVERSION OF SYNTHETIC METHOSTENOL-4-C^{14} TO CHOLESTEROL[1]

Sir:

We have reported recently that sodium acetate-1-C^{14} injected into rats intracardially becomes incorporated into the methostenol (4α-methyl-Δ^7-cholesten-3β-ol) of the skin and liver–small in-

(1) This investigation was supported by a research grant (H-2458-C3) from the National Institutes of Health, Public Health Service.

testine pool.[2] Specific activity findings suggested a precursor relationship between methostenol and cholesterol. Methostenol-4-C^{14} was prepared in order to subject this hypothesis to experimental test. Cholesterol-4-C^{14} [3] (0.05 mC.) diluted with 2.0 g. of cholesterol served as starting material. Methostenol-4-C^{14} was prepared by first synthesizing 7-dehydrocholesterol-4-C^{14} according to the method of Hunziker and Müllner.[4] Hydrogenation of 7-dehydrocholesterol-4-C^{14} in dioxane over Raney nickel[5] afforded Δ^7 - cholestenol - 4 - C^{14}. Methostenol-4-C^{14} then was prepared from Δ^7-cholestenol-4-C^{14} by methods previously described.[6,7] After repeated chromatography on silicic acid: Celite (2:1) columns, methostenol-4-C^{14}, m.p. 143–5°, exhibited a specific activity of 9,650 c.p.m./mg. A solution of radioactive methostenol in Tween-80[8] was fed by stomach tube to two male albino rats (A and B) of the Holtzman strain, weighing 155 and 110 gm., respectively. The former received 89,700 c.p.m. and was sacrificed 6 hours after administration of the meal and the latter received 134,000 c.p.m. and was sacrificed 24 hours after intubation. A control was prepared which consisted of the liver and small intestine of a similar rat to which was added 4,000 c.p.m. of methostenol-4-C^{14} and sufficient alcoholic KOH to saponify the mixture. At each time of sacrifice, liver–intestine pool, carcass, and feces were separately saponified with alcoholic KOH. The non-saponifiable fraction of each preparation was isolated with ether, taken to dryness and liver–intestine and carcass mixtures were chromatographed on silicic acid: Celite (2:1) columns (1.8 × 20 cm.) using an elution gradient of benzene:Skelly C (2:1) against a reservoir of Skelly C. Fractions were plated and radioactivity measured with a thin end window gas flow counter. Results of the recovery of radioactivity in the methostenol or cholesterol fractions are shown in Table I. Specific activity values for cholesterol were determined after several crystallizations from methanol and a purification through the dibromide.[9] Melting points were 146–148°.

TABLE I

THE RADIOACTIVITY OF CERTAIN STEROLS ISOLATED AFTER THE INGESTION OF A METHOSTENOL-4-C^{14} MEAL BY THE RAT[a]

Rat no.	Tissue	Recovery of C^{14} in non-sap. fraction: Total c.p.m.	Methostenol c.p.m.	Cholesterol c.p.m.	Cholesterol specific activity c.p.m./mg.
Control	Liver–Int.	3,750	3630	120	0
A (6 hr.)	Liver–Int.	14,500	4900	6170	199
	Carcass	5,400	1760	3040	39 9
B (24 hr.)	Liver–Int.	4,024	1270	2530	116
	Carcass	3,840	630	2950	21.7

[a] Control received 4,000 c.p.m.; rat A, 89,700 c.p.m.; rat B, 134,000 c.p.m.

(2) C. L. Lorah and W. W. Wells, *Fed. Proc.*, **18**, 276 (1959).
(3) Nuclear-Chicago Corp.
(4) F. Hunziker and F. X. Müllner, *Helv. Chim. Acta*, **41**, 70 (1958).
(5) L. F. Fieser and J. E. Herz, THIS JOURNAL, **75**, 121 (1953).
(6) W. W. Wells and D. H. Neiderhiser, *ibid.*, **79**, 6569 (1957).
(7) D. H. Neiderhiser and W. W. Wells, *Arch. Biochem. & Biophys.*, **81**, 300 (1959).
(8) Polyoxyethylene sorbitan monooleate, Atlas Powder Co.
(9) E. Schwenk and N. T. Werthessen, *Arch. Biochem. Biophys.*, **40**, 334 (1952).

A large fraction of the recovered C^{14} was located in the saponifiable fraction of the feces (rat A, 28.6%; rat B, 85.7%) presumably as bile acids and their derivatives. The rapid conversion of methostenol to cholesterol places methostenol in the role of an active intermediate of cholesterol biosynthesis. The recent isolation and characterization of 4α-methyl-Δ^8-cholesten-3β-ol from mice preputial gland tumors[10] suggests the interesting possibility that this sterol could give rise to methostenol by the migration of the double bond at C-8 (9) to C-7, *in vivo*.

(10) A. A. Kandutsch and A. E. Russell, THIS JOURNAL, **81**, 4114 (1959).

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RECEIVED SEPTEMBER 30, 1959

π-COMPLEXES OF THE TRANSITION METALS. XII. SYNTHESIS WITH ALKYL CHROMIUM AND NICKEL COMPOUNDS

Sir:

One-step syntheses of polynuclear benzenoid hydrocarbons, *i.e.*, naphthalenes, phenanthrenes and anthracenes, by cyclic condensation of disubstituted acetylenes on triarylchromium(III) compounds have been reported.[1] In these reactions it is clear that the aryl groups bonded to chromium have participated in the cyclizations and substituted hydrogen has been concurrently abstracted from them. Although diarylnickel(II) derivatives are quite capable of cyclizing acetylenes to the benzene ring system, they do not form polynuclear aromatic structures.[2] We conclude, therefore, that organochromium(III) is a powerful *hydrogen acceptor*. This conclusion, in experimental trial, has led to unique syntheses which we now report.

Triethylchromium(III) is prepared by the same general method employed in the synthesis of the corresponding phenyl derivative.[3] In tetrahydrofuran this alkylchromium compound also cyclizes tolane to hexaphenylbenzene; but also, *it contributes an ethyl group* in a mixed condensation with tolane, yielding *1,2,3,4-tetraphenylbenzene*, I, m.p. 187–189°.[4] Thus, organochromium(III), in addition to providing a two-carbon atom fragment for condensation with two molecules of tolane, has further demonstrated its hydrogen acceptor capacity by dehydrogenation of the dihydrobenzene ring.

Diethylnickel in tetrahydrofuran also condenses tolane to hexaphenylbenzene and provides an ethyl group for a mixed condensation. However, the relatively weak hydrogen accepting characteristic of organonickel(II) is manifested by reaction termination at the *dihydro stage*, and *1,2,3,4-tetraphenyl-1,3-cyclohexadiene*, II, m.p. 170–171° [*Anal.* Calcd. for $C_{30}H_{24}$: C, 93.71; H, 6.29.

(1) H. Zeiss and W. Herwig, THIS JOURNAL, **80**, 2913 (1958); W. Herwig, W. Metlesics and H. Zeiss, *ibid.*, **81**, Dec. 5 (1959).
(2) M. Tsutsui and H. Zeiss, Abstracts, 134th Meeting, Amer. Chem. Soc., Chicago, Sept., 1958, p. 59-P.
(3) W. Herwig and H. Zeiss, THIS JOURNAL, **81**, 4798 (1959).
(4) Identification was made by physical and spectroscopic comparisons with authentic substance.