is to be expected that deviations from Henry's law would apparently reduce the non-uniformity of the solute.

Summary

The theory for the determination of average molecular weights for mixtures has been developed and the need for distinguishing various kinds of "averages" has been discussed. It has been shown how number-average, weight-average, and Z-average molecular weights may be calculated from data on sedimentation equilibrium in the Svedberg ultracentrifuge, and the methods have been applied to experimental data on gelatin. A numerical measure for the non-uniformity of mixtures with respect to molecular weight has been proposed. The significance of the results in connection with macromolecular materials, such as cellulose, rubber, proteins, etc., has been explained and the correct method for comparing molecular weights by osmotic pressure or equivalent methods and Staudinger's viscosity method has been elucidated.

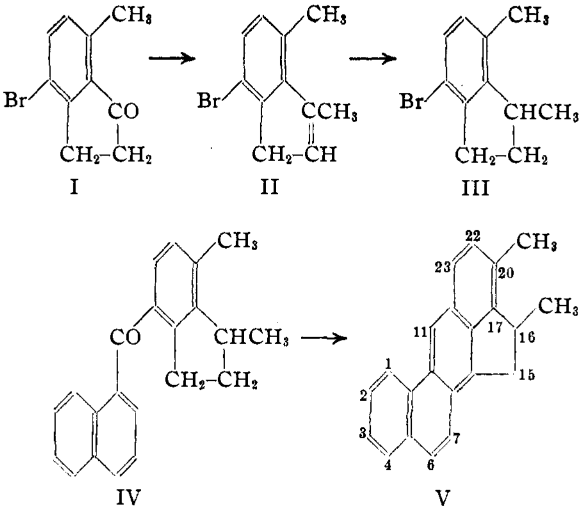
WILMINGTON, DELAWARE RECEIVED APRIL 8, 1935

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF HARVARD UNIVERSITY]

16,20-Dimethylcholanthrene

BY LOUIS F. FIESER AND ARNOLD M. SELIGMAN

With the idea that optically active substances capable of producing cancer in test animals would afford a very interesting field for investigation, we have as a first step synthesized a hydrocarbon, V, having an asymmetric carbon atom and of a type likely to possess carcinogenic activity. Starting with 7-methyl-4-bromohydrindone-1 (I)[1] the synthesis was accomplished through the series of transformations I ⟶ V, the ketone IV being obtained by the interaction of the Grignard reagent from III with α-naphthoyl chloride. The very faintly yellow hydrocarbon resulting from the pyrolysis of IV has the composition and properties consistent with formula V[1a]. Since the compound is a homolog of the carcinogenically active[2] methylcholanthrene, and since it would be expected from the work of Barry and others[3] that the branching produced by the added methyl group in the aliphatic chain attached to the 1,2-benzanthracene nucleus at the favorable C_5-position would enhance the potency, the new hydrocarbon may be a favorable case for study. Tests for activity are being made with the *dl*-compound before attempting a resolution.



Since the name "cholanthrene" has been applied[4] to the unsubstituted pentacyclic structure, it is convenient to refer to the new hydrocarbon as a dimethylcholanthrene. To provide a system of numbering capable of application both to cholanthrene and to its hydro-derivatives, we propose to use the sterol numbering without modification, even though this has the awkward feature of assigning the numbers 20 and 22 to ortho positions in a benzene ring. According to the suggested system (see formula V), the carcinogenic hydrocarbon obtained from bile acids acquires the specific name of 20-methylcholanthrene and the new hydrocarbon is the 16,20-dimethyl derivative of the parent hydrocarbon.

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[1] Fieser and Seligman, THIS JOURNAL, **57**, 942 (1935).

[1a] Dr. Egon Lorenz reports that the absorption spectrum of the hydrocarbon is practically identical in intensity and in the positions of the bands with that of methylcholanthrene. The absorption spectrum of cholanthrene, the parent hydrocardon recently synthesized by Cook, Haslewood and Robinson [*J. Chem. Soc.*, 667 (1935)], is also of a similar pattern; as compared with methylcholanthrene, the bands are somewhat sharper and there is a general shift of about 10 Å. in the direction of shorter wave length. A synthesis of cholanthrene by the modified Elbs reaction will be reported shortly.

[2] Cook and Haslewood, *J. Chem. Soc.*, 428 (1934).

[3] Barry, Cook, Haslewood, Hewett, Hieger and Kennaway, *Proc. Roy. Soc.* (London), **B117**, 318 (1935).

[4] Wieland and Dane, *Z. physiol. Chem.*, **219**, 240 (1933).

Experimental Part

1,7-Dimethyl-4-bromoindene-1 (II).—A Grignard reaction, using 25 g. of 7-methyl-4-bromohydrindone-1, 32 g. of methyl iodide, and 8.3 g. of magnesium, and refluxing the mixture for fifteen minutes, gave, after evaporation of the washed and dried ethereal extract, 26 g. of the crude carbinol as a viscous oil. This was heated at 190° for twenty minutes to effect dehydration, and the indene was purified by steam distillation from a mixture of the somewhat dark oil with dilute sodium hydroxide solution. The colorless, mobile oil was extracted, dried and distilled in vacuum; b. p. 110–111° at 1.8 mm.; yield, 15.6 g. (63%). The substance decomposes when distilled at atmospheric pressure and it polymerizes on long standing.

Anal. Calcd. for $C_{11}H_{11}Br$: C, 59.19; H, 4.98. Found: C, 59.30; H, 4.61.

1,7-Dimethyl-4-bromohydrindene (III) was obtained in 80% yield by hydrogenating the unsaturated compound in absolute alcohol solution using Adams catalyst. After removing the solvent by fractionation, the product was obtained as a mobile liquid, b. p. 98.5° at 1.5 mm.

Anal. Calcd. for $C_{11}H_{13}Br$: C, 58.67; H, 5.82. Found: C, 58.61; H, 5.50.

1,7-Dimethyl-4-(α-naphthoyl)-hydrindene (IV).—The Grignard reagent from 12 g. of III was prepared by the technique described in our earlier work[1] and added to a stirred solution of 23 g. of α-naphthoyl chloride in ether at −10°. After refluxing overnight and decomposing the mixture, the ether-soluble portion was subjected to steam distillation in the presence of alkali in order to remove an oily by-product (dimethylhydrindene?) and to hydrolyze unchanged acid chloride. After extracting with ether and thorough washing with alkali, the reaction product was distilled in vacuum, giving a viscous, pale yellow oil; yield, 9.4 g. (59%). Seed was obtained after standing for several weeks, and the substance then crystallized from petroleum ether as small, colorless needles melting at 112–114°.

Anal. Calcd. for $C_{22}H_{20}O$: C, 87.95; H, 6.72. Found: C, 87.87; H, 6.77.

16,20-Dimethylcholanthrene (V).—Pyrolysis of the crude ketone (9 g.) under nitrogen at 400–405° for thirty minutes gave, after distillation in vacuum and one crystallization from ether, 1.7 g. (20%) of yellow hydrocarbon, m. p. 154–155°. The material was purified through the picrate and crystallized once from ether: yellow, m. p. 161–162°. A solution in benzene (50 cc. per 0.1 g.) was passed through an adsorption tower of activated alumina, which removed further colored impurities: pale yellow plates, m. p. 166–167.5°, corr. After four repetitions of the adsorption process, the hydrocarbon was obtained as plates from ether, m. p. 169–170°, corr., having a barely visible yellow tinge. The solution in concentrated sulfuric acid is orange-red. On mixing the sample with methylcholanthrene the melting point was depressed about 14°.

Anal. Calcd. for $C_{22}H_{18}$: C, 93.58; H, 6.43. Found: C, 93.58; H, 6.35.

The *picrate* crystallizes from benzene as black prisms melting at 179–180°, corr. (depression with methylcholanthrene picrate).

Anal. Calcd. for $C_{28}H_{21}O_7N_3$: N, 8.22. Found: 8.33.

CONVERSE MEMORIAL LABORATORY
CAMBRIDGE, MASSACHUSETTS
RECEIVED JUNE 18, 1935

NOTE

The Oxidizing Faculty of Nitrogen Oxide

BY JANNIK BJERRUM[1] AND LEONOR MICHAELIS

It has been recently observed by Meyerhof[2] that in certain types of biological systems which utilize molecular oxygen as oxidant in the respiration process, nitric oxide can be used in place of oxygen, being reduced to nitrous oxide during the process. This observation suggested that nitric oxide might be reduced by other easily oxidizable organic compounds. The experiment was tried with various leuco dyes, such as those of methylene blue, pyocyanine and 2,6-chloroindophenol, with substituted *p*-phenylenediamines, and with semiquinoid violet reduction products of the quaternary γ,γ'-dipyridylium compounds (viologens).

The dye was dissolved in a buffer solution, some drops of a 1% solution of colloidal palladium added, hydrogen bubbled through the system until the dye was reduced, the hydrogen washed out with nitrogen and nitric oxide generated from ferrous sulfate and sodium nitrite, according to the procedure of O. Warburg,[3] was passed through the solution. The dye was gradually oxidized. With methylene blue this was shown to occur both in acid and alkaline solution, in aqueous solution and in alcoholic solution.

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(1) Fellow of the Rockefeller Foundation.

(2) Meyerhof and Schulz, *Biochem. Z.*, **275**, 147 (1934).

(3) O. Warburg, *ibid.*, **189**, 354 (1927).