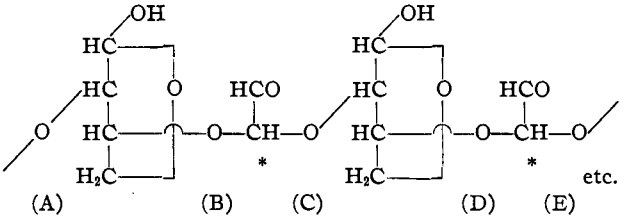
[CONTRIBUTION FROM THE RESEARCH LABORATORY OF ORGANIC CHEMISTRY, MASSACHUSETTS INSTITUTE OF TECHNOLOGY, NO. 259]

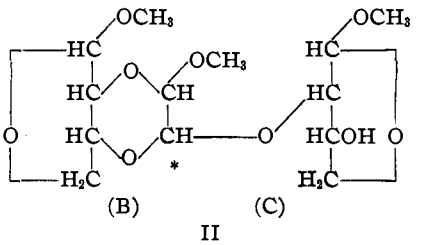
The Reaction between Periodate Oxidized Starch and Methanol Containing Hydrogen Chloride

BY J. H. MICHELL[1] AND C. B. PURVES

When the tendency of erythrose to assume a furanoid form is remembered,[2] the structure accepted for starch oxidized by one mole of periodic acid[3] can be written as I. The glyoxal acetal bonds A, C, E, etc., are the glycosidic bonds of the original starch while B, D, etc., are the original pyranoside ring formations. Methanol containing hydrogen chloride would cleave these two types of bonds in random order and would also methylate the reducing groups. Each cleavage would produce a new hydroxyl group in either the second or third position of an erythrose residue and the formation of a hemiacetal with the free aldehyde group of the neighboring glyoxal unit would result in a substituted 1,4-dioxane ring. The probability of such a cyclization is strengthened by the assumption of similar, although unstable, structures for the dimeric forms of glycol aldehyde and glyceric aldehyde[4,5] and by the existence of well-defined, crystalline substances such as that prepared by warming glyoxal sulfate with ethylene glycol. This substance probably consisted of two 1,4-dioxane rings fused together in the 2,3-position.[6] If scission first took place at A and was followed in order by cyclization between the erythrose unit AB and the glyoxal residue BC, by methylation of hemiacetal hydroxyl groups and by a final rupture at D, the structure II would result. Initial rupture at D and a final methanolysis at A would produce the isomeric fragment III in similar fashion. Equal amounts of II and III would not be expected because the bonds A and D are of different types and might not undergo methanolysis at quite the same rate. One of the two possibilities, indeed, might not materialize at all. The presence of methylglycosidic groups, rather than unsubstituted hemiacetals, in II and III, should make these structures stable enough to isolate and characterize.[7] The methanolysis of starch oxidized by periodate did in fact result in a 2% yield of crystals, m. p. 148°, which agreed in composition and properties with either of these two formulas.[8] Further experiments compatible with the ideas just outlined for the course of the methanolysis are described in the present article.



I



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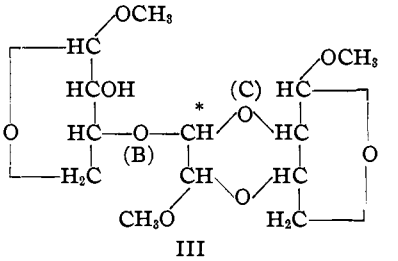


III

Experimental

Distillation of the Product from the Methanolysis of Starch Oxidized by Periodate.—The preparation of a chloroform soluble sirup from 41.4 g. of the oxidized starch was previously described.[8] Residual chloroform mixed

(1) Abstracted from a thesis submitted by J. H. Michell to the Faculty of the Massachusetts Institute of Technology in partial fulfillment of the requirements for the degree of Doctor of Science, 1941.

(2) Hockett and Maynard, THIS JOURNAL, **61**, 2111 (1939).

(3) Jackson and Hudson, *ibid.*, **60**, 989 (1938).

(4) Fischer and Baer, *Ber.*, **63**, 1744 (1930); **65**, 337, 345 (1932), and other articles. See also Wohl and Neuberg, *ibid.*, **33**, 3095 (1900).

(5) Bergmann and Miekeley, *ibid.*, **62**, 2297 (1929), and earlier articles.

(6) Baker and Field, *J. Chem. Soc.*, 86 (1932).

(7) Butler and Cretcher, THIS JOURNAL, **54**, 2987 (1932), heated 2,3-dichlorodioxane in absolute alcohol and obtained a 71% yield of the 2,3-diethoxy derivative, b. p. 95–98° (14 mm.). Böeseken, Tellegen and Henriquez, *ibid.*, **55**, 1284 (1933), mention the surprising stability of dioxane 2,3-diacetate, m. p. 104–105°, toward dilute acid or alkali and that of a hexachlorodioxane, m. p. 91°, toward alcoholic silver nitrate or caustic potash.

(8) Michell and Purves, THIS JOURNAL, **64**, 585 (1942).

with a little glyoxal tetramethylacetal passed into the receiver when this sirup was fractionally distilled,[9] b. p. 31–50° (3 mm.). The remainder, 31 g., gave a low-boiling fraction, b. p. 110–134° (3–4 mm.), $n^{20}D$ 1.4510 (9.1 g.), an intermediate portion (1.5 g.) and a high-boiling fraction; b. p. 185–227° (3–15 mm.) (9.1 g.). Decomposition set in toward the end of the distillation and the residue (9.3 g.) was a black solid. The course of the distillation (Fig. 1, plot I) showed clearly that only two main fractions were present.

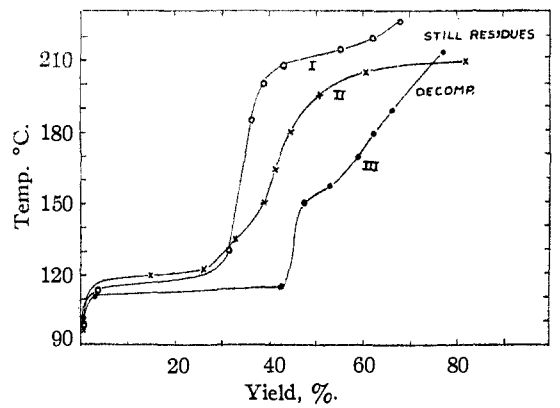


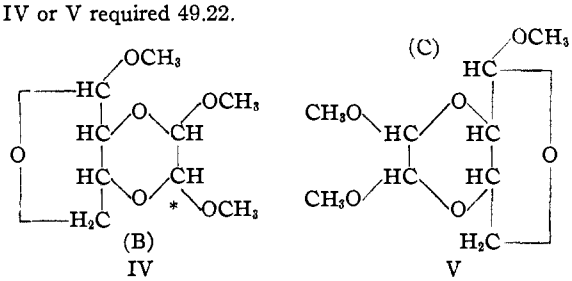
Fig. 1.—Distillation of methanolysis products at 3–4 mm. pressure: Plot I, periodate oxidized starch; Plot II, high boiling fraction from periodate starch; Plot III, periodate oxidized cellulose.

Examination of the High-Boiling Fraction.—Partial crystallization occurred when this fraction (9.1 g.) was kept overnight at 50° and after trituration with ether a filtration removed the crystals to which the alternative structures II and III were attached.[8] The yield after purification to the correct melting point of 148° was 0.75 g. or 9% of the entire fraction. Redistillation of the uncrystallized portion (7.7 g.) between the limits 195–205° (3 mm.) mostly occurred from 202–204° and the distillate was a deep yellow glass at 25°, an extremely viscous oil at 80°. It dissolved readily in most organic solvents but only sparingly in water. The specific levorotation for an 0.966% aqueous solution was −53.6°[10]

Anal. Calcd. for structure II or III, $C_{10}H_{12}O_5(OCH_3)_3$: C, 48.4; H, 6.9; OCH_3, 28.9; mol. wt., 322. Found: C, 48.8, 48.7; H, 7.4, 7.2; OCH_3, 32.3, 32.5.

An ebullioscopic molecular weight determination with an 85.0 mg. sample in 15.47 g. of carbon tetrachloride[11] gave 320. In an estimation of glyoxal units,[8] a 233.1 mg. sample gave 27.5 mg. of crude glyoxal-*bis*-2,4-dinitrophenylhydrazone or 90% of the theoretical for structures II and III.

Examination of the Low-Boiling Fraction.—About 95% passed into the receiver between 116 and 119.5° when this fraction was redistilled, at 5 mm., through a Podbielniak column. The distillate was a non-reducing, slightly viscous, water-white liquid with a slight odor like that of acetal. It dissolved freely in all solvents tried except petroleum ether and the specific levorotation in 0.6% aqueous solution was −90.5°. The molecular weight of 226 in carbon tetrachloride,[11] together with the specific gravity (20°/20°) of 1.204 and the refractive index, $n^{20}D$ 1.4488, gave a molecular refraction of 49.05. Structure IV or V required 49.22.



A 3.63% solution of the substance was made by heating on the steam-bath for fifteen minutes with decinormal hydrochloric acid, and, after cooling, the specific levorotation was −87.9°. Further heating for one and one-half and for two hours changed the levorotation to −51.3° and −46.4° but the gradual formation of a finely divided brown precipitate then made the solution too dark to read. The latter rotation corresponded to one of −85° on the assumption that one mole of erythrose had been formed from structure IV or V. As *d*-erythrose probably has a specific rotation much less levo than −85°,[12] the result was anomalous.

On standing overnight, the redistilled low-boiling fraction solidified to a white, waxy texture and 3 g. was dissolved in 20 ml. of an ether–petroleum ether (3:10) mixture. After keeping at 0° for two days, 0.57 g. of a white, bulky precipitate separated and was recrystallized from petroleum ether. The specific levorotation was −59.1° (0.22% aqueous solution) and a portion, sublimed in high vacuum at 60°, melted at 97–98° (cor.).

Anal. Calcd. for structure IV or V, $C_6H_7O_5(OCH_3)_3$: C, 49.1; H, 7.3; OCH_3, 42.3; mol. wt., 220. Found: C, 49.1, 49.0; H, 7.5, 7.4; OCH_3, 41.5, 41.6.

A 54.5 mg. sample in 10 ml. (15.81 g.) of carbon tetrachloride gave mol. wts. of 216 and 209.[11] In an estimation of glyoxal units,[8] 119.4 mg. gave 28.1 mg. of the crude *bis*-2,4-dinitrophenylhydrazone, or 99% of the amount calculated for structures IV or V.

The above crystalline material amounted to about 18% of the low-boiling fraction. After removal of solvent, the remaining 80% was redistilled under diminished pressure. The distillate was a clear colorless liquid; b. p. 117–118° (4 mm.), $n^{20}D$ 1.4489; sp. gr.$^{20}_{20}$, 1.209, with a specific levorotation of −91.7° in 0.781% aqueous solution.

Anal. Calcd. for structure IV or V, $C_6H_7O_5(OCH_3)_3$: C, 49.1; H, 7.3; OCH_3, 42.3; mol. wt., 220. Found: C, 49.1, 48.8; H, 7.6, 7.5; OCH_3, 41.6, 42.3.

A 163.2-mg. sample in 15.72 g. of carbon tetrachloride gave mol. wts. of 223 and 222.

A 116.2-mg. sample gave 30.0 mg. of crude glyoxal-*bis*-2,4-dinitrophenylhydrazone or 110% of the amount calculated for III or IV. The molecular refraction of 48.85 checked theory of 49.22.

Degradation of Liquid High-Boiling Isomers.—After the removal of the crystalline portion, 16 g. of the liquid

(9) Morton, "Laboratory Technique in Organic Chemistry," McGraw-Hill Book Company, New York, N. Y., 1938, p. 78.

(10) Optical rotations were observed at 20° in sodium light.

(11) Swietoslawski, *Bull. soc. chim.*, **49**, 1563 (1931).

(12) Felton and Freudenberg, THIS JOURNAL, **57**, 1637 (1935).

residues, b. p. 195–205° (3 mm.), were boiled under reflux for three hours with 133 g. of a 10% solution of hydrogen chloride in dry methanol. The black brown solution was worked up by methods already indicated[8] and the sirup extracted from aqueous solution by chloroform was distilled to remove a small amount of glyoxal tetramethylacetal, b. p. 30–50° (3–4 mm.). Fractionation of the residue, 8.5 g., gave two distinct fractions (Fig. 1, Curve II) of which the one boiling at 150–220° (3–4 mm.) on redistillation (4.7 g.) was disregarded as being essentially the original substance. The fraction boiling between 120–135° weighed only 2.7 g. and was not redistilled. Its specific levorotation in 0.902% aqueous solution was −73.2° and $n^{20}D$ was 1.4505.

Anal. Calcd. for structure IV or V, $C_6H_7O_3(OCH_3)_3$: C, 49.1; H, 7.3; OCH_3, 42.3. Found: C, 48.7, 48.3; H, 7.6, 7.7; OCH_3, 41.5, 42.6.

The yield of this fraction, calculated on the amount of the original material actually degraded (11.3 g.) was about 25% by weight or about 36%.

A More Limited Methanolysis of Oxidized Starch.—The periodate oxidized starch had been kept over calcium chloride for two months. A 20-g. sample dissolved almost completely when heated under reflux for one hour with 245 g. of 1% hydrogen chloride in dry methanol. The excess solvent was distilled from the neutralized, brown solution and the residue was steam distilled to remove glyoxal tetramethylacetal. A dark, heavy tar, soluble in warm alcohol, insoluble in chloroform, and solidifying to an apparently crystalline mass separated. The chloroform extracts of the aqueous residue were concentrated to 300 ml. and the addition of an equal volume of ether precipitated a brown mass (4 g.) fusing between 155–160° without becoming completely liquid and not apparently changed by a further three-hour treatment with 1% acid methanol. The mother liquors from this precipitate were concentrated to 50 ml. and the addition of 200 ml. of ether gave a second precipitate which after isolation resolved itself into a brown oil and a white powder. The former (6.4 g.) exuded much ether when dried at 50°, became a semi-crystalline mass, fusing at 130–140°, and gave a very viscous liquid, insoluble in water, on retreatment for three hours with 1% acid methanol. The powder, after reprecipitation from chloroform–ether, fused between 140 and 160°. Its complete insolubility in ether showed that it was not identical with crystalline fractions produced in the much more drastic methanolysis of starch.

Although the above experiment was a preliminary one and was complicated by tar formation during the steam distillation, the products had the qualitative behavior of mixtures of polymers of varying molecular weight. The gradation in solubility and the tendency to form micro crystals with indefinite melting points in the same range, lent support to this view.[13]

The Action of Hydrogen Chloride in Methanol on Cellulose Oxidized with Periodate.—Purified cotton linters, 32.4 g. or 0.2 mole, was oxidized at 20° with 54.4 g. or 0.2 mole of sodium paraperiodate buffered to *p*H 4.1 as already described for starch.[8] The periodate in the liquor disappeared after four days of constant agitation and the yield of oxidized cellulose was only 28.0 g. This low yield suggested much secondary oxidation.

The 28 g. of oxy-cellulose was heated under reflux for five hours in 10% methanol–hydrogen chloride and the products were isolated as in the similar work with starch, the only change being that ether instead of chloroform was used in the extraction of the aqueous solution. The residual oil from the ether extract, 4.0 g., on distillation at 3–4 mm., pressure gave a fraction, 1.7 g., b. p. 112–115°. Thereafter the temperature rose sharply to 150° and a second fraction (about 1 g.) distilled with decomposition between 150 and 220° (Fig. 1, plot III). This fraction corresponded to the higher boiling product from periodate oxidized starch but trituration with cold ether failed to separate a similar crystalline portion. The refractive index of the lower boiling fraction (1.7 g.) changed from $n^{20}D$ 1.4460 to $n^{20}D$ 1.4466 on redistillation but an accidental loss prevented further purification. An 0.942% aqueous solution had a specific levorotation of −88.1°.

Anal. Calcd. for structure IV or V, $C_6H_7O_3(OCH_3)_3$: C, 49.1; H, 7.2; OCH_3, 42.3. Found: H, 48.8, 48.2; H, 6.7, 7.6; OCH_3, 41.1, 40.9.

A 128.0-mg. sample gave 32.4 mg. of crude glyoxal-*bis*-2,4-dinitrophenylhydrazone or 134%. The fraction was obviously not quite pure.

Discussion

Inspection of the formulas II and III shows that each should occur in eight isomeric forms corresponding to the various possible alpha and beta configurations of the three methoxyl groups. The high boiling fraction from the methanolysis of the oxidized starch gave one of these isomers in a pure crystalline state.[8] Although the residual glass was somewhat discolored by traces of volatile decomposition products from the residue in the still, elementary analysis, methoxyl content and molecular weight estimations also agreed with II and III. The structural identity of the glassy portion and the crystals was strongly supported by the high yield of combined glyoxal recovered from the former. It followed that the entire high boiling fraction from the methanolysis consisted of two or more isomers like II or III. As the combined yield represented about 22% of the starch oxidized, alternative C_3–C_2 structures for the crystals were definitely eliminated.

In addition to the high boiling fraction, a mobile, colorless fraction $n^{20}D$ 1.4488, distilled sharply at 116–119° (3–4 mm.) (Fig. 1, plot I). The fraction separated into a pure crystalline substance, m. p. 97–98°, $[\alpha]^{20}D$ −59.1° in water, and a liquid residue with $n^{20}D$ 1.4489 and a specific rotation of −91.7°. These two fractions both agreed with the molecular formula $C_6H_7O_3(OCH_3)_3$ and both gave glyoxal tetramethylacetal on further meth-

(13) Carothers and Hill, THIS JOURNAL, **54**, 1559 (1932).

anolysis. The entire low boiling fraction from the oxidized starch was therefore a mixture of isomers. Considerations outlined in the introduction suggested that the mixture consisted of two or more of the isomeric hexahydro-2,3,5-trimethoxyfuro-[3,4]-*p*-dioxins (IV and V). Structure IV was readily obtained from that of oxidized starch (I) by the preferential rupture of the bonds A, C, E, etc., while V arose from scission at B, D, etc. If the starred carbon atoms retained the original alpha glycosidic configuration of starch, the structures had a pair of alpha and beta methylglycosidic isomers in common and the total number of isomers was six. If the asymmetry of the starred carbon atom was variable, IV and V represented two out of eight possible forms. Attempts to discriminate between the possibilities by the use of model formulas suggested that all isomers were stereochemically possible, although it appeared that the 1,4-dioxane ring could exist only in the "bath" form and never in the "chair" form when derived from a glucopyranoside.

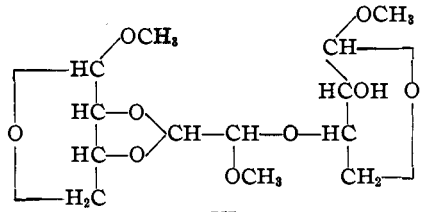
In another experiment, the high-boiling fraction (II) or (III) was submitted to incomplete degradation in acid methanol. On distillation, the product included some unchanged material (Fig. 1, plot II) and a new low-boiling fraction. Although the small scale of the preparation precluded complete purification of the latter and it did not partially crystallize, analytical and other data left no doubt that II or III had been degraded to IV or V in about 36% of the theoretical yield. This conversion was entirely in keeping with the proposed structures and directly proved the C_4–C_2–C_4 carbon skeleton of the original material.

Another deduction from theory was that structures analogous to II and IV, but of greater molecular weight, formed in the initial stages of the methanolysis of oxidized starch (I) by the preferential scission of widely spaced bonds like A and F. A methanolysis was carried out with 1% instead of 10% acid methanol and much of the product had the expected solubility in organic solvents. Although crystalline or semi-crystalline fractions were obtained, they could not be purified to solids of narrow melting range and the work was discontinued. The still residues from the distillation of the products obtained in the more drastic methanolysis (Fig. 1, plot I) were possibly of a similar nature. If they had the constitution just suggested for them, they corresponded to about 27% of the periodate oxidized starch, while another 42% was recovered as the high and low boiling fractions. These figures accounted for about 57% of the erythrose and 69% of the glyoxal residues. The rest of the glyoxal could be recovered as the tetramethylacetal and the missing erythrose was either destroyed during the methanolysis or was not extracted by chloroform from the aqueous solution of the product.

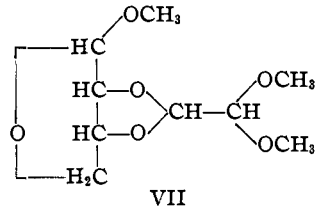
Although the crystals II or III consumed the proper amount of oxidant in a simultaneous hydrolysis and oxidation with aqueous periodic acid, erythrose could not be positively identified in a hydrochloric acid hydrolyzate. The final specific rotation, based on erythrose, was +81° in this case[8] and −85° for the liquid mixture IV or V. Such widely discordant values suggest that the glyoxal and erythrose initially formed had recombined to a variable but considerable extent under the influence of the aqueous acid. Cyclic acetals are readily obtained from phenylglycol and aliphatic aldehydes,[14] or from bromoacetaldehyde and mannitol,[15] in similar conditions. A recondensation of glyoxal and erythrose would explain the surprisingly low yields isolated from the hydrolysis of periodate oxy-starch with aqueous acid.[3]

The fact that periodate oxy-cellulose behaved similarly[16] to the oxy-starch in methanolysis proved that various branched chain structures considered for starch[17–20] were not necessary for the production of the derivatives II to V. The latter must therefore be connected either with the degradation of linear chains of anhydroglucose units or with the recondensation of glyoxal and erythrose in acid methanol. Although the production of II to V, especially II or III, in the observed yields by any reversion process seems improbable, the possibility was not definitely excluded by the present work. In this event the products of the methanolysis may not have the 1,4-dioxane structures tentatively assigned to them but may be cyclic acetals of types VI and VII. Cyclization of carbonyl with conveniently situated hydroxyl groups, together with the pos-

(14) Verley, *Bull. soc. chim.*, [3] **22**, 275 (1899).
(15) Hibbert and Hill, THIS JOURNAL, **45**, 734 (1923).
(16) As cellulose is a beta and starch an alpha glucoside, the carbon atoms starred in structures II to V may have opposite configurations. In this event the products from cellulose would be stereoisomers of those from starch.
(17) Meyer, *Helv. Chim. Acta*, **23**, 845 (1940).
(18) Staudinger and Huseman, *Ann.*, **527**, 195 (1936).
(19) Hirst and Young, *J. Chem. Soc.*, 1471 (1939).
(20) Tilden and Hudson, THIS JOURNAL, **61**, 2900 (1939).



VI



VII

sible recondensation of initial scission products, are therefore factors which must be simultaneously considered during research in the oxy-starch and oxy-cellulose field.

The authors are indebted to Drs. E. J. Crane, L. T. Capell and A. M. Patterson for help in naming the acetals II to V.

Summary

1. The distillation, at 3–4 mm., of the chloroform soluble products from the methanolysis of starch oxidized by periodate gave two fractions, b. p. 116–119° and 195–205°, in yields of about 18% and 24%, respectively.

2. Detailed chemical study showed that the higher-boiling fraction was probably a mixture of isomers in which a methylerythroside unit was combined in a dioxane ring with a glyoxal unit, to which in turn a methoxyl group and a second methylerythroside group were attached. These isomers were hexahydro-3,5-dimethoxy-2-(1-methyl-3, or 2-erythrofuranosyloxy)furo[3,4]-*p*-dioxins.

3. The low-boiling fraction was considered to be a mixture of isomers containing a methylerythroside residue condensed in a dioxane ring with a glyoxal residue to which two methoxyl groups were attached. The structures were those of the isomeric hexahydro-2,3,5-trimethoxyfuro[3,4]-*p*-dioxins and one isomer was isolated in a pure condition, m. p. 97–98°, $[\alpha]^{20}D$ −59° in water.

4. When the higher boiling fraction was partially degraded by methanolysis, a 36% yield of a lower boiling fraction resulted and the properties of this fraction were those described in section (3).

5. The methanolysis of periodate oxidized cellulose took a course similar to that described in sections (1) to (3) for starch.

6. The data were consistent with the assumption that the acetal links in long, uniform chains of the oxidized starch underwent methanolysis in random order while dioxane rings resulted from the formation of new hemiacetal bonds. If, however, the data depended on the recondensation of glyoxal and erythrose in acid methanol, five-membered cyclic acetal structures were not excluded for the substances described in (2) to (5). Preference was given to the former alternative.

7. The suggestion was made that glyoxal and erythrose recondensed to an unknown extent in the presence of aqueous acid.

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Steric Influences on the Aromaticity of Dipyrrylmethenes. The Synthesis and Study of the Properties of a Di-N-methyldipyrrylmethene[1]

BY KARL J. BRUNINGS AND ALSOPH H. CORWIN

The preparation of N-methyl pyrroles and N-methyl dipyrrylmethanes as the first step in the synthesis of N-methylated porphyrins has been described in the first paper of this series.[2] The condensation of these compounds according to standard methods failed to provide the expected dipyrrylmethenes, which are intermediates in the proposed porphyrin synthesis. In those cases where the expected methene was to have both nitrogens occupied by methyl groups, red oils were obtained which could not be identified and the reactions which should have yielded unsymmetrical mono-N-methylmethenes provided instead totally unsubstituted dipyrrylmethenes.[3,4,5] The subsequent study of these reactions suggested that methylated methenes were unstable.

By a change in the technique of isolation we have been able to prepare 1,3,5,1′,3′,5′-hexa-

(1) Studies in the Pyrrole Series VI. Paper V, Corwin and Krieble, THIS JOURNAL, **63**, 1829 (1941).

(2) Corwin and Quattlebaum, *ibid.*, **58**, 1081 (1936).

(3) Corwin and Andrews, *ibid.*, **58**, 1086 (1936).

(4) Corwin and Andrews, *ibid.*, **59**, 1973 (1937).

(5) Paden, Corwin and Bailey, *ibid.*, **62**, 418 (1940).