It was slightly resinified by boiling with potassium hydroxide solution, but it failed to reduce Fehling's solution even upon continued boiling.

The quantity was not large enough to determin satisfactorily whether a sulfite compound was formed or not, and the attempt to prepare a small quantity of the semicarbazone was unsuccessful.

It is dextrorotatory.

An aldehyde with the formula $C_{12}H_{20}O$, with two ethylene linkages, would be an olefin, homologous with geraniol. The name *cypral* will be given to this compound, as at present no such homolog of the olefinic camphors is known.

As circumstances have made necessary the postponement of this work for several months, the details so far gathered have been given in this paper for the interest of those engaged in terpene investigations.

Summary.

The summary of the physical properties of the two new compounds described is as follows:

Cypressene, $C_{15}H_{24}$, a yellowish-green, viscous, and almost odorless oil, b. p. 218-220°$_{35}$ and 295-300°$_{778}$. Sp. gr. $0.9647^{18}/_{4}$. n_D^{22} 1.5240. $[\alpha]_D^{20} = 6.53°$. *Cypral*, $C_{12}H_{20}O$, a light yellow, mobil, and very fragrant oil of b. p. 182-185°$_{35}$. Sp. gr. $0.9469^{20}/_{4}$. n_D^{20} 1.5040. Dextrorotatory.

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[CONTRIBUTIONS FROM THE SHEFFIELD LABORATORY OF YALE UNIVERSITY.]

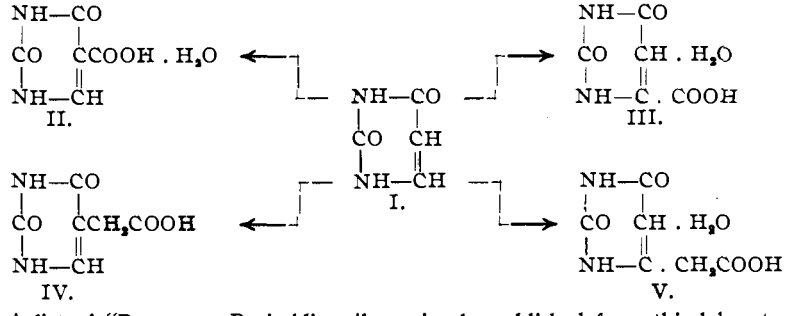
RESEARCHES ON PYRIMIDINES: SYNTHESIS OF CYTOSINE-5-ACETIC ACID.

[FIFTY-FIRST PAPER.][1]

BY TREAT B. JOHNSON (Experimental work by HARLEY T. PECK and JOSEPH A. AMBLER).

Received *March 21*, 1911.

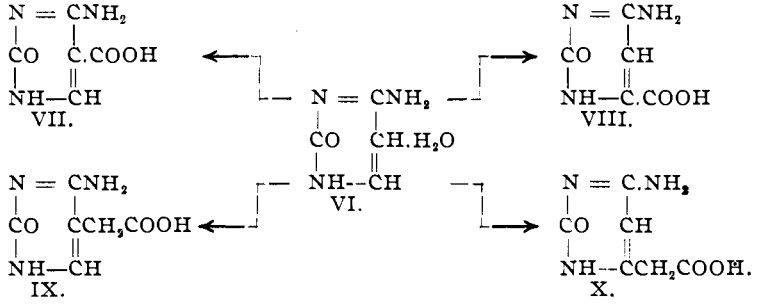
If one considers only the carbon-substitution products of uracil (I), there are two possible uracilcarboxylic and two uracilacetic acids. They are represented by the structural formulas, II, III, IV, and V.



[1] A list of "Papers on Pyrimidines," previously published from this laboratory, is appended to this paper.

Three of these have been synthesized in this laboratory, *viz.:* uracil-5-carboxylic acid[1] (II), uracil-5-acetic acid[2] (IV) and uracil-4-acetic acid[3] (V). The last member of the series, or uracil-4-carboxylic acid (III), was prepared by Müller[4] and its constitution later definitly established by Wheeler.[5]

Of the four corresponding acids of cytosine (VI), only one has previously been described, *viz.:* cytosine-5-carboxylic acid (VII), which was synthesized by Wheeler and Johns.[6] A description of the synthesis and properties of cytosine-5-acetic acid (IX) will be given in this paper.



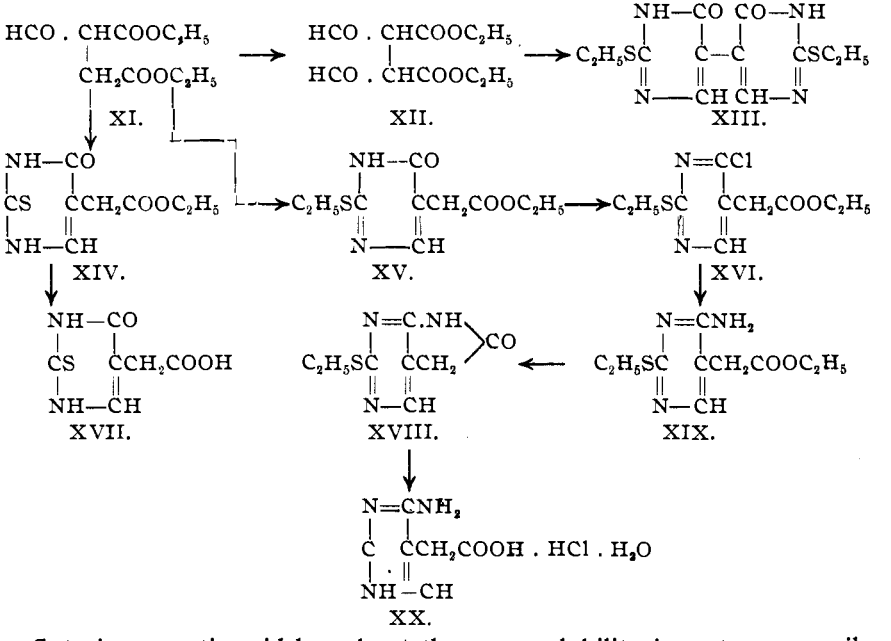
Johnson and Speh[7] have shown that the sodium salt of diethyl formylsuccinate (XI) condenses smoothly with pseudoethylthiourea, in aqueous solution, giving ethyl 2-ethylmercapto-6-oxypyrimidine-5-acetate (XV). We now find that this mercaptopyrimidine (XV) reacts with phosphorus oxychloride under proper conditions, without destruction of the ester grouping, giving an excellent yield of ethyl 2-ethylmercapto-6-chlorpyrimidine-5-acetate (XVI). Especially interesting was the behavior of this chloride towards ammonia. The corresponding aminopyrimidine (XIX) was not obtained as expected, but it underwent an inner condensation giving smoothly the γ-lactam of 2-ethylmercapto-6-amino-pyrimidine-5-acetic acid (XVIII), with formation of a molecule of alcohol. This cyclic compound underwent hydrolysis easily, by heating with concentrated hydrochloric acid, giving practically a quantitative yield of the hydrochloride of cytosine-5-acetic acid (XX). The free acid was then obtained by treatment of this salt with one molecular proportion of alkali, in aqueous solution.

Thiourea and diethyl formylsuccinate condensed smoothly, in the presence of sodium ethylate, giving a good yield of ethyl 2-thio-6-oxypyrimi-

[1] Wheeler, Johnson and Johns, *Am. Chem. J.*, **37**, 392.
[2] Johnson and Speh, *Ibid.*, **38**, 602.
[3] Wheeler and Liddle, THIS JOURNAL, **30**, 1156.
[4] *J. prakt. Chem.*, **56**, 488.
[5] *Am. Chem. J.*, **38**, 358.
[6] *Ibid.*, **38**, 595.
[7] *Loc. cit.*

dine-5-acetate (XIV). When this was saponified with alkali the corresponding acetic acid (XVII) was obtained.

Diethyl succinate apparently can condense with two molecular proportions of ethyl formate giving the disodium salt of diethyl diformylsuccinate (XII). The yield however is very small. Its formation was shown by the fact that the condensation product of pseudoethylthiourea and sodium diethylformylsuccinate was, in two experiments, a mixture of ethyl 2-ethylmercapto-6-oxypyrimidine-5-acetate (XV), and a difficultly soluble substance having the composition of the dipyrimidine (XIII). Enough of the latter pyrimidine however was not obtained for a thorough examination. These various transformations are represented by the following formulas:



Cytosine-5-acetic acid has about the same solubility in water as uracil, thymine or uracil-5-acetic acid (IV). Like its lower homolog, cytosine-5-carboxylic,[1] it crystallizes from water in an anhydrous condition. Cytosine,[2] on the other hand, crystallizes with one molecule of water. This property of these cytosine acids to crystallize from water in an anhydrous condition is worthy of attention. Uracil crystallizes from water in anhydrous form while, on the other hand, every acid of the series represented above, *viz.*: (II), (III), (IV) and (V), crystallizes with one molecule of water, with the exception of uracil-5-acetic acid (IV). Cytosine-5-

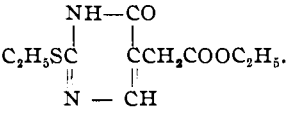
[1] *Loc. cit.*

[2] Weeler and Johnson, *Am. Chem. J.*, **29**, 492.

acetic acid is not precipitated from an aqueous solution of its hydrochloride by phosphotungstic acid, while cytosine is practically quantitatively precipitated with this reagent. The acid like cytosine-5-carboxylic acid[1] forms a characteristic monohydrochloride, which crystallizes with one molecule of water.

Experimental Part.

Ethyl 2-ethylmercapto-6-oxypyrimidine-5-acetate,



This compound has been described in a previous paper from this laboratory.[2] It is easily prepared by condensing pseudoethylthiourea with the sodium salt of diethyl formylsuccinate in aqueous solution.

The sodium salt was prepared by condensing 45 grams of ethyl formate with 100 grams of diethyl succinate in the presence of 13 grams of sodium, suspended in 150 cc. of anhydrous ether. After the condensation was complete the salt was then dissolved in 600 cc. of cold water and the solution divided into two equal parts. Assuming that the yield of sodium salt was theoretical (129 grams), a molecular proportion of pseudoethylthiourea hydrobromide (53 grams), dissolved in 100 cc. of water, was added to the first half of the salt solution, and 27 grams of the hydrobromide, in 100 cc. of water, added to the second portion. Molecular proportions of potassium hydroxide (16 and 8 grams) were then finally added to combine with the hydrobromic acid and the two solutions allowed to stand, at ordinary temperature, for 12 hours and finally heated for half an hour at 100°. After cooling and acidifying the solutions with glacial acetic acid, we obtained from the first fraction in which molecular proportions were used 30 grams and from the second part 18 grams of the mercaptopyrimidine. In another experiment, in which more than one molecular proportion of the pseudourea hydrobromide was used, the yield was not increased. The experiments demonstrate therefore that the yield of pyrimidine is decreased by using less than one molecular proportion of the hydrobromide for a condensation. A yield of 30 grams of the mercaptopyrimidine from 50 grams of diethyl succinate corresponds to 43.2 per cent. of the theoretical.

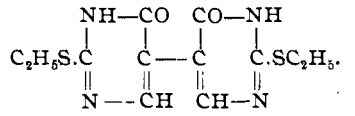
In one experiment 20 grams of diethyl succinate were condensed with 17 grams of ethyl formate (2 mols.) in presence of 6.0 grams of sodium suspended in ether. After standing for 5 days the crude salt was dissolved in 125 cc. of water and 42 grams of the hydrobromide of pseudoethylthiourea, dissolved in 75 cc. of water, added to the solution. Thirteen

[1] *Loc. cit.*

[2] Johnson and Speh, *Loc. cit.*

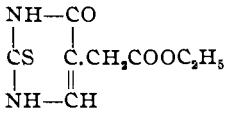
grams of potassium hydroxide, in 20 cc. of water, were then added and the solution allowed to stand for 13–14 hours. After heating half an hour to complete the reaction, cooling and acidifying with acetic acid, we obtained a heavy deposit of ethyl 2-ethylmercapto-6-oxypyrimidine-5-acetate mixed with a small amount of a fine crystallin powder. This was separated by trituration of the crude reaction product with warm alcohol. The compound was insoluble in this solvent and water and difficultly soluble in acetic acid. It contained sulfur and had no definit melting point but charred and decomposed on heating. It was purified for analysis by repeated digestion with glacial acetic acid. Nitrogen determinations agreed with the calculated value for

2_2-Diethylmercapto-6_2-dioxy-5,5-dipyrimidine,



Calculated for $C_{12}H_{14}O_2N_4S_2$: N, 18.06; found, 17.70, 17.95.

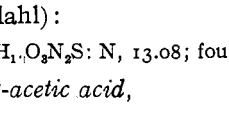
Ethyl 2-thio-6-oxypyrimidine-5-acetate,



The sodium salt of diethyl formylsuccinate was prepared in the usual manner by condensation of 20 grams of diethyl succinate with 9 grams of ethyl formate in presence of 2.6 grams of sodium. After dissolving in 120 cc. of cold water, 8 grams of thiourea, in 75 cc. of water, were added to the solution and the mixture allowed to stand about 10 hours. It was then heated on the steambath for half an hour, cooled and acidified with glacial acetic acid. Eight grams of the thiopyrimidine separated as a yellow crystallin powder. The pyrimidine was purified by crystallization from hot water and separated in prismatic crystals melting at 178–180°. Analysis (Kjeldahl):

Calculated for $C_8H_{10}O_3N_2S$: N, 13.08; found, 13.09, 13.04.

2-Thio-6-oxypyrimidine-5-acetic acid,

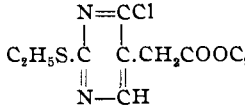


The potassium salt of this acid was obtained by saponification of the preceding ethyl ester with aqueous potassium hydroxide. When the alkaline solution was acidified with hydrochloric acid this pyrimidine separated in prismatic crystals. The yield was quantitative. It is purified

best by crystallization from hot water and melts with decomposition at $\pm 260°$, according to the rate of heating. Analysis (Kjeldahl):

Calculated for $C_6H_6O_3N_2S$: N, 15.05; found, 14.83, 15.00.

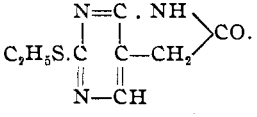
Ethyl 2-ethylmercapto-6-chloropyrimidine-5-acetate,



Fifteen grams of ethyl 2-ethylmercapto-6-oxypyrimidine-5-acetate were dissolved in 50 cc. of phosphorus oxychloride and digested for half an hour when the evolution of hydrochloric acid practically ceased. The excess of chloride was then removed by distillation under diminished pressure when we obtained a sirupy liquid, which was poured upon crushed ice to decompose any phosphorus halide present. The pyrimidine separated as an oil insoluble in water and was dissolved in ether and dried over calcium chloride. It boiled at 220° at 31 mm. pressure. The yield of purified oil was 7 grams or about 44 per cent. of theory. In two other experiments, in which we used 30 grams of the mercaptopyrimidine and 75 cc. of the phosphorus halide, we obtained 15 and 14 grams of the chloride boiling at 203–203.5° at 16 mm. and 215° at 28 mm. pressure respectively. Analysis (Kjeldahl):

Calculated for $C_{10}H_{13}O_2SCl$: N, 10.74; found, 11.0, 10.50.

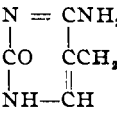
γ-Lactam of 2-ethylmercapto-6-aminopyrimidine-5-acetic acid or 2-ethylmercapto-5,6-(α-pyrrolidone)-pyrimidine,



This pyrimidine was obtained, in an attempt to prepare ethyl 2-ethylmercapto-6-aminopyrimidine-5-acetate, by heating ethyl 2-ethylmercapto-6-chloropyrimidine-5-acetate with ammonia. Fifteen grams of the 6-chloropyrimidine were dissolved in 100 cc. of strong alcoholic ammonia and heated for 5 hours at 120–130°. When the tube was opened a crystallin substance was suspended in the alcohol mixed with ammonium chloride. The alcohol was removed by evaporation on the steambath and the product obtained triturated with cold water to dissolve the ammonium chloride. The undissolved pyrimidine was then dissolved in hot alcohol and the solution cooled, when it separated as a red granular powder which melted at 208°. The yield was good. The compound is difficultly soluble in water and soluble in acids. Analysis (Kjeldahl):

Calculated for $C_{10}H_{16}O_2N_3S$: N, 17.42.
Calculated for $C_8H_9ON_3S$: N, 21.54.
Found: I, 21.57; II, 21.62.

Cytosine-5-acetic acid,



This acid was obtained in the form of its hydrochloride by hydrolysis of the above γ-lactam with concentrated hydrochloric acid. The salt crystallizes from 20 per cent. hydrochloric acid in minute needles, which decompose at 135–140° with effervescence, and contain one molecule of water of crystallization, $C_6H_7O_3N_3.HCl.H_2O$. Water determination:

Calculated for $C_6H_7O_3N_3.HCl.H_2O$: H_2O, 8.05; found, 8.3.

Chlorine determination in hydrous salt:

Calculated for $C_6H_7O_3N_3.HCl.H_2O$: Cl, 15.88; found, 15.95, 16.04.

Cytosine-5-acetic acid was obtained by dissolving this hydrochloride in the required volume of 0.1 *N* sodium hydroxide solution and heating to boiling temperature. On cooling, the acid deposited in granular, orange-colored crystals. After decolorization with bone-coal the acid is colorless and blackens in a capillary tube at 240–250°, but does not melt below 290°. The acid was dried for analysis in a desiccator over calcium chloride. It does not contain water of crystallization. One hundred grams of water dissolve at 25° 0.3373 gram of the acid. Analysis (Kjeldahl):

Calculated for $C_6H_7O_3N_3$: N, 24.85; found, 24.62, 25.01, 24.86.

Cytosine-5-acetic acid is not precipitated from an aqueous solution of its hydrochloride by addition of phosphotungstic acid, mercuric chloride or copper sulfate. Potassio-bismuth iodide gives an amorphous precipitate which dissolves in an excess of hydrochloric acid.

Picrate.—Crystallizes from hot water in clusters of needles, which melt at 217–218° to a dark red oil.

Addenda.

For the sake of reference, a complete list of the papers on pyrimidines, which have been published from this laboratory, is given below. These papers are arranged in the order in which they appeared.

1. Researches on the Cycloamidines: Pyrimidine Derivatives. By Henry L. Wheeler. *Am. Chem. J.*, 20, 481.

2. On Some Condensation Products of the Pseudothioureas: Synthesis of Uracil, Thymine and Similar Compounds. By Henry L. Wheeler and Henry F. Merriam. *Am. Chem. J.*, 29, 478.

3. Synthesis of Amino-oxypyrimidines Having the Composition of Cytosine: 2-Amino-6-oxypyrimidine and 2-Oxy-6-aminopyrimidine. By Henry L. Wheeler and Treat B. Johnson. *Am. Chem. J.*, 29, 492.

4. On Cytosine or 2-Oxy-6-aminopyrimidine from Triticonucleic Acid. By Henry L. Wheeler and Treat B. Johnson. *Am. Chem. J.*, 29, 505.

5. 5-Methylcytosine. By Henry L. Wheeler and Treat B. Johnson. *Am. Chem. J.*, 31, 591.

6. Synthesis of 2-Amino-5-methyl-6-oxypyrimidine. By Treat B. Johnson and Samuel H. Clapp. *Am. Chem. J.*, 32, 130.

7. 2-Oxy-4,6-Diaminopyrimidine. By Henry L. Wheeler and George S. Jamieson. *Am. Chem. J.*, **32**, 342.

8. The Structure of Some Substitution Products. By Henry L. Wheeler and H. Stanley Bristol. *Am. Chem. J.*, **33**, 437.

9. The Action of Potassium Thiocyanate upon Some Imide Chlorides. By Henry L. Wheeler and H. Stanley Bristol. *Am. Chem. J.*, **33**, 448.

10. The Action of Aqueous and Alcoholic Ammonia and Aniline on Some Halogen and Mercapto Pyrimidines. By Treat B. Johnson and Carl O. Johns. *Am. Chem. J.*, **34**, 175.

11. 2-Ethylmercapto-5-amino-6-oxypyrimidine. By Treat B. Johnson. *Am. Chem. J.*, **34**, 191.

12. On 2,5-Diamino-6-oxypyrimidine. By Treat B. Johnson and Carl O. Johns. *Am. Chem. J.*, **34**, 554.

13. Some 5-Iodopyrimidine Derivatives: 5-Iodocytosine. By Treat B. Johnson and Carl O. Johns. *J. Biol. Chem.*, **1**, 305.

14. On Methods of Synthesizing Isobarbituric Acid and 5-Oxycytosine. By Treat B. Johnson and Elmer V. McCollum. *J. Biol. Chem.*, **1**, 437.

15. The Action of Potassium Thiocyanate upon Imide Chlorides. By Treat B. Johnson and Elmer V. McCollum. *Am. Chem. J.*, **36**, 136.

16. On the Formation of Purines from Ureapyrimidines. By Treat B. Johnson and Elmer V. McCollum. *Am. Chem. J.*, **36**, 149.

17. On 5-Nitrocytosine and its Reduction to 2-Oxy-5,6-diaminopyrimidine. By Treat B. Johnson, Carl O. Johns and Frederick W. Heyl. *Am. Chem. J.*, **36**, 160.

18. 5-Ethylcytosine. By Treat B. Johnson and George A. Menge. *J. Biol. Chem.*, **2**, 105.

19. Synthesis of Uracil-5-carboxylic Acid. By Henry L. Wheeler, Treat B. Johnson and Carl O. Johns. *Am. Chem. J.*, **37**, 392.

20. On a Color Test for Uracil and Cytosine. By Henry L. Wheeler and Treat B. Johnson. *J. Biol. Chem.*, **3**, 183.

21. On Some Salts of Cytosine, Isocytosine, 6-Aminopyrimidine and 6-Oxypyrimidine. By Henry L. Wheeler. *J. Biol. Chem.*, **3**, 285.

22. Uracil-4-carboxylic Acid. By Henry L. Wheeler. *Am. Chem. J.*, **38**, 358.

23. The Action of Methyl Iodide on 2-Anilino-6-oxypyrimidine, and the Synthesis of 2-Anilinopyrimidine. By Treat B. Johnson and Frederick W. Heyl. *Am. Chem. J.*, **38**, 237.

24. Synthesis of Thymine-4-carboxylic Acid. By Treat B. Johnson. *J. Biol. Chem.*, **3**, 299.

25. Synthesis of Cytosine-5-carboxylic Acid. By Henry L. Wheeler and Carl O. Johns. *Am. Chem. J.*, **38**, 594.

26. Synthesis of Thymine-5'-carboxylic Acid. By Treat B. Johnson and Carl Frank Speh. *Am. Chem. J.*, **38**, 602.

27. Synthesis of 4-Methyluracil-5-acetic Acid. By Treat B. Johnson and Frederick W. Heyl. *Am. Chem. J.*, **38**, 659.

28. A Method of Separating Thymine from Uracil. By Treat B. Johnson. *J. Biol. Chem.*, **4**, 407.

29. The Action of Nitric Acid on 2,6-Dioxypyrimidines. Oxynitrohydrothymine. By Treat B. Johnson. *Am. Chem. J.*, **40**, 19.

30. Synthesis of Uracil-3-acetic Acid. By Henry L. Wheeler and Lenoard M. Liddle. THIS JOURNAL, **30**, 1152.

31. Synthesis of Uracil-4-acetic Acid. By Henry L. Wheeler and Leonard M. Liddle. THIS JOURNAL, **30**, 1156.

32. Synthesis of Some Nitrogen Alkyl Derivatives of Cytosine, Thymine and Uracil. By Treat B. Johnson and Samuel H. Clapp. *J. Biol. Chem.*, **5**, 49.

33. The Action of Diazo-Benzene Sulfonic Acid on Thymine, Uracil and Cytosine. By Treat B. Johnson and Samuel H. Clapp. *J. Biol. Chem.*, **5**, 163.

34. The Action of Potassium Thiocyanate upon Some Imide Chlorides. By Treat B. Johnson and Walter F. Storey. *Am. Chem. J.*, 40, 131.

35. Synthesis of Cytosine-5-carboxamide. By Henry L. Wheeler and Carl O. Johns. *Am. Chem. J.*, 40, 233.

36. Synthesis of 4-Methylcytosine. By Carl O. Johns. *Am. Chem. J.*, 40, 348.

37. On Some Picrolonates. By Henry L. Wheeler and George S. Jamieson. *J. Biol. Chem.*, **4**, 111.

38. Synthesis of Some Benzyl Derivatives of Uracil and Thymine. By Treat B. Johnson and John H. Derby, Jr. *Am. Chem. J.*, 40, 444.

39. Synthesis of New Derivatives of 5-Hydroxyuracil (Isobarbituric Acid). By Treat B. Johnson and D. Breese Jones. *Am. Chem. J.*, 40, 538.

40. The Thio Derivatives of Uracil and the Preparation of Uracil in Quantity. By Henry L. Wheeler and Leonard M. Liddle. *Am. Chem. J.*, 40, 548.

41. On the Formation of Purine Derivatives from 4-Methylcytosine. By Carl O. Johns. *Am. Chem. J.*, 41, 58.

42. Synthesis of 1-Methyl-5-hydroxyuracil. By Treat B. Johnson and D. Breese Jones. THIS JOURNAL, **31**, 591.

43. The Preparation of 3-Methyl- and 3-Benzyluracil. By Henry L. Wheeler and Treat B. Johnson. *Am. Chem. J.*, 42, 30.

44. The Preparation of 1,4-Dimethyluracil and of the Monobenzyl derivatives of 4-Methyluracil. By Henry L. Wheeler and David F. McFarland. *Am. Chem. J.*, 42, 101.

45. Sulphur Derivatives of 5-Hydroxyuracil: Preparation of 5-Benzylmercaptouracil and 5-Benzylmercaptocytosine. By Treat B. Johnson and Herbert H. Guest. *Am. Chem. J.*, 42, 271.

46. Dimethyl Derivatives of 2–Aminopyrimidine, Preparation of 2-Methylamino-5-methylpyrimidine. By Treat B. Johnson and Kenneth G. Mackenzie. *Am. Chem. J.*, 42, 353.

47. The Action of Methyl Iodide, and of Benzyl Chloride upon 2-Oxy-4-methyl-6-mercaptopyrimidine. By Henry L. Wheeler and David F. McFarland. *Am. Chem. J.*, 42, 431.

48. Synthesis of 5-Cyanuracil. By Treat B. Johnson. *Am. Chem. J.*, 42, 505.

49. The Thio Derivatives of Thymine and the Preparation of Thymine. By Henry L. Wheeler and David F. McFarland. *Am. Chem. J.*, 43, 19.

50. The Condensation of Thiourea with Esters of Allylmalonic and Some Alkyl Substituted Allylmalonic Acids. By Treat B. Johnson and Arthur J. Hill. *Am. Chem. J.*, 45, 356.

NEW HAVEN, CONN.

[CONTRIBUTION FROM THE HAVEMEYER LABORATORIES OF COLUMBIA UNIVERSITY, NO. 189.]

A TETRA-ACETYL AMINOGLUCOSIDE.

BY MARSTON LOVELL HAMLIN.

Received March 27, 1911.

A preliminary report of work on chitin begun independently last October, suggested in part by investigations of Offer[1] as to the structure of chitin,

[1] *Biochem. Z.*, **7**, 117.