characteristic of azo dyes formed by the coupling of diazonium salts with β -naphthol, the diazo group being alpha to the hydroxyl. Dropped on heated porcelain, the compound puffs suddenly in dense, red fumes and on combustion leaves no ash.

Ten g. skeins of silk, wool and cotton were dyed by developing the color directly in the fibre, by passing the wet skeins first through a bath containing 0.5 g. of the diazonium chloride and one cc. conc. hydrochloric acid in 300 cc. water, and then through a solution of 0.2 g. β -naphthol and one g. sodium hydroxide in 300 cc. water. The silk and cotton were dyed salmon pink, the wool a bright orange without exhausting the baths. The color proved fast to water, soap, dilute acid and light.

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[CONTRIBUTION FROM THE SHEFFIELD CHEMICAL LABORATORY OF YALE UNIVERSITY.]

RESEARCHES ON PYRIDINES. LXXXVIII. THE SYNTHESIS OF CYTOSINE ALDEHYDE.

BY TREAT B. JOHNSON AND LOUIS A. MIKESKA.

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Comparatively little is known about pyrimidine aldehydes. Uracil-aldehyde (I), the first of the series of these interesting compounds to be described, was prepared by Johnson and Cretcher¹ and the only other representative known, namely, thymine-aldehyde (II), was prepared by the same investigators somewhat later.² In order to extend our knowledge of this class of compounds we decided to prepare cytosine-4-aldehyde (III), and the object of this paper is to give a description of a method of synthesis which gives promise of enabling us to prepare this combination in quantity. Our work, however, has not been brought to a satisfactory conclusion and this paper therefore deals only with the chemistry of the preliminary reactions involved in the process.

As the starting point of a cytosine aldehyde synthesis we selected the acetal of 2-ethylmercapto-6-oxy-4-aldehydopyrimidine (V), which was first described by Johnson and Cretcher,³ and subjected this to the action of phosphorus halides (POCl_3 and PCl_5) in order to obtain the imide chloride or its chloro-ether as represented by Formulas VI and IV, respectively. To our surprise the product of the reaction proved to be the true

¹ THIS JOURNAL, 37, 2144 (1915).

² J. Biol. Chem., 26, 99 (1916).

³ Loc. cit.

aldehyde corresponding to Formula VIII. It is remarkable that this combination is formed as a product of this reaction as most acetals give chloro-ethers on treatment with phosphorus halides. It made no difference whether the reaction product was poured into water or into an alkaline solution to destroy the unaltered phosphorus halide after completion of the reaction; in every case the free aldehyde was obtained. The results obtained seemed to indicate that the acetal grouping forms some complex with the phosphorus oxychloride which can be decomposed only to the aldehyde, or that the aldehyde group which results from this reaction is stable in the presence of this reagent. To settle this question 2-ethylmercapto-6-chloro-4-aldehydopyrimidine (VIII) was treated with an excess of phosphorus oxychloride under conditions similar to those which were employed for its preparation, and, indeed, the aldehyde was recovered unaltered.

Ammonia reacts normally with the chloropyrimidine (VIII) replacing the chlorine atom by an amino group. The reaction, however, does not proceed smoothly and does not stop with the displacement of the halogen atom. Three distinct compounds were isolated as products of this reaction. The ammonia reacted not only with the halogen but also with the aldehyde group giving 2-ethylmercapto-6-amino-4-imidomethylpyrimidine (VII) which was easily separated from the other substances on account of its insolubility in cold alcohol. This pyrimidine theoretically should yield by hydrolysis cytosine-4-aldehyde (XI). The limited amount of the substance available did not permit a thorough investigation of its behavior towards hydrolytic agents, but some evidence was obtained however, which seemed to indicate that the hydrolysis does not stop with the formation of cytosine-4-aldehyde (XI), but that the compound is reduced completely to uracil. If our preliminary observation is confirmed by further investigations, which is now in progress, the result may throw some light upon the possible structure of the natural nucleosides.

Another substance which was obtained as a reaction product of 2-ethylmercapto-6-chloro-4-aldehydopyrimidine (VIII) and alcoholic ammonia was the anhydro derivative of 2-ethylmercapto-4-aldehydo-6-amino-pyrimidine represented by Formula IX. This combination undoubtedly results from the inner condensation of the corresponding aldehyde (X), a behavior which is not surprising in view of the fact that similar condensations are known to take place in the aromatic series. Anhydro-*o*-aminobenzaldehyde and the corresponding *meta* compound are representatives of this type.¹

We did not succeed in isolating and purifying the 2-ethylmercapto-6-amino-4-aldehydopyrimidine (X). It represented apparently the chief

¹ Posner, *Ber.*, 31, 653 (1898); Beilstein's *Handbuch*, 3, 12 (*D. R. P.* 62950).

product of the reaction, and when digested with hydrochloric acid was converted into cytosine-4-aldehyde (XI) with evolution of ethyl mercaptan. These various transformations are expressed by the following formulas:

While the synthesis outlined above has enabled us to prepare the hitherto unknown cytosine-aldehyde (XI) it has not been perfected to such a degree as to be recommended for the preparation of this interesting product in large quantities. Further work is in progress dealing with a study of the action of aqueous ammonia on the chloropyrimidine (VIII) and also the conditions influencing the formation of the various possible intermediate products.

Experimental Part.

Ethyl γ -Diethoxy-acetoacetate, $(\text{C}_2\text{H}_5\text{O})_2\text{CH.COCH}_2\text{COOC}_2\text{H}_5$.—This ester has been prepared previously by Dakin and Dudley¹ and later by Johnson and Cretcher² by the action of sodium on a mixture of ethyl diethoxyacetate and ethyl acetate. We found it advisable to introduce a slight modification of their methods in order to obtain the best possible yields. A description of one experiment will illustrate our method of preparation together with the changes introduced. The following proportions were taken, namely, 180 g. of the diethoxyacetate, 48 g. of metallic sodium (2 mols) in wire form and 277 g. of ethyl acetate (3 mols). It will be noted that only two molecular proportions of sodium were used whereas the previous workers used three. The diethoxy ester and $1/2$ of the ethyl acetate were mixed in a round bottom flask and the mixture heated to 80° . While maintaining the temperature at 80° , $1/2$ of the

¹ *J. Chem. Soc.*, **105**, 2453 (1914).

² *THIS JOURNAL*, **37**, 2144 (1915).

sodium was added in small amounts, all of which dissolved in about 2 hours. The remainder of the ethyl acetate was then added and finally, in small amounts, the remainder of the sodium. The heating was continued for 3 hours when the sodium had all dissolved. The red viscous mass was then poured into enough cold water to dissolve the salts and the cooled solution acidified directly with hydrochloric acid. The β -ketone ester was extracted with ether, and, after thorough drying over sodium sulfate and removal of the ether, it was obtained as a red oil. The ethyl acetoacetate and unchanged esters were removed by heating the crude oil to 140° under a pressure of 19 mm. We had left after this treatment 202 g. of the crude ketone ester, which was used for condensations without further purification.

This undistilled ketone ester was condensed with thiourea according to the method of Johnson and Cretcher¹ and converted into the acetal of 2-thio-4-uracilaldehyde. The proportions employed for the synthesis of this pyrimidine were as follows: 150 g. of the crude β -ketone ester, 17 g. of sodium, 65 g. of thiourea and 325 cc. of absolute alcohol. After 7 hours' digestion to complete the condensation the excess of alcohol was removed by heating under reduced pressure and the residue obtained dissolved in 300 cc. of water. This solution was then decolorized by means of bone charcoal and finally acidified with hydrochloric acid when the pyrimidine separated. This was purified by crystallization from alcohol and melted at 160° .

Diethylacetal of 2-Ethylmercapto-6-oxy-4-aldehydopyrimidine,

the method already described by Johnson and Cretcher.¹ From 110 g. of the corresponding 2-thiopyrimidine we obtained 105 g. of this mercapto compound. This corresponds to a yield of 85% of the theoretical.

The Action of Phosphorus Halides on the Acetal of 2-Ethylmercapto-6-oxy-4-aldehydopyrimidine.

Ten g. of the pyrimidine acetal and 20 cc. of phosphorus oxychloride were heated for 2 hours at $75-80^\circ$. Hydrochloric acid gas was evolved during this operation and the solution itself assumed a yellowish brown color. On pouring into cold water to decompose the excess of phosphorus

¹ *Loc. cit.*

halide the chloropyrimidine separated in the form of a dark-colored oil. This was extracted with ether, dried over calcium chloride and then purified by distillation under diminished pressure. It boiled at $138-139^{\circ}$ under a pressure of 10 mm. The yield was excellent.

Calc. for $C_7H_7ON_2ClS$: N, 13.86; Cl, 17.55. Found: N, 13.92; Cl, 17.34.

In one experiment we removed the excess of phosphorus oxychloride by distillation under reduced pressure, and then poured the crude reaction product into a cold dil. aqueous solution of sodium hydroxide. The solution was kept cold by adding crushed ice and allowed to stand for about 2 hours before extracting the oil with ether. We obtained the chloropyrimidine after removal of the ether, in the form of a dark-colored oil. It was purified by distillation and boiled at $151-158^{\circ}$ under a pressure of 14 mm. and after a second distillation completely solidified on cooling.

Attempts to prepare this compound by heating the pyrimidine acetal with phosphorus oxychloride at 140° were unsuccessful. Much resinous material was produced by such treatment and practically no chloropyrimidine was obtained. With phosphorus pentachloride the acetal reacted smoothly to give the chloropyrimidine if the reaction mixture was not heated above 80° . The formation of this pyrimidine by the action of this phosphorus halide is quite remarkable in that the aldehyde group is not destroyed.

Phenylhydrazone of 2-Ethylmercapto-6-chloro-4-aldehydopyrimidine, $C_6H_5N_2SCl.CH = NNHC_6H_5$.—A quantitative yield of this hydrazone is obtained when the aldehyde described is combined with phenylhydrazine in ether solution. On evaporating the ether it separated in a crystalline condition and was purified for analysis by crystallization from ligroin. It crystallized in the form of long, yellow needles which melted at 147° . This compound was extremely soluble in all the common organic solvents, but was insoluble in water.

Calc. for $C_{13}H_{13}N_4SCl$: Cl, 12.14. Found: 12.10.

The Anil of 2-Ethylmercapto-6-chloro-4-aldehydopyrimidine, $C_6H_5N_2SCl.CH = NC_6H_5$.—This base was prepared by mixing the pyrimidine aldehyde (10 g.) with aniline (4.5 g.). They reacted at ordinary temperature with evolution of heat giving a crystalline product which was very soluble in ether. For analysis, the anil was purified by crystallization from 50% alcohol. It separated from this solvent in the form of badly distorted prisms which melted at 85° to a yellow oil. The anil was very soluble in all the common organic solvents but insoluble in water.

Calc. for $C_{13}H_{12}N_2SCl$: N, 16.00. Found: 16.06.

The Action of Ammonia on 2-Ethylmercapto-6-chloro-4-aldehydopyrimidine.—Several experiments were conducted before conditions were

established whereby it became possible for us to isolate in a pure condition the different products of reaction which are formed when ammonia interacts with this chloride. While the aldehyde group was not altered by the action of phosphorus halides, on the other hand, it was very susceptible to the action of ammonia, and for this reason we had to deal with a property which very much complicated our problem. In fact, the experimental procedures that enabled us to obtain cytosine combinations in previous work could not be applied successfully in the present case on account of the various secondary reactions which took place.

We shall record, therefore, only the results of a single experiment which is representative of a series, and was productive of three characteristic products of reaction.

Eight g. of 2-ethylmercapto-6-chloro-4-aldehydopyrimidine was heated under pressure with an excess of a saturated solution of ammonia in alcohol. On mixing these two reagents there is no apparent reaction, but the pyrimidine dissolves completely at ordinary temperature giving a solution which is intensely colored and strongly fluorescent. By reflected light the solution was blue bordering on violet, while by transmitted light it was reddish brown. The mixture was heated for two hours at 119-124° and the solution then allowed to cool, when two distinct substances separated in a crystalline form; one was identified as ammonium chloride and the other as 2-ethylmercapto-6-amino-4-imidomethylpyrimidine,

The contents of the bomb tube was filtered and the ammoniacal filtrate saved (see below). The insoluble material containing ammonium chloride was washed with cold water to remove this salt when we obtained the *imide* in a very pure condition. It was purified for analysis by crystallization from absolute alcohol, and separated from this solvent in the form of prisms which melted at 182° without decomposition. The pyrimidine was insoluble in alkali, but was very soluble in acids as would be expected from its constitution. The yield of this substance was about 3.0 g.

Calc. for $\text{C}_7\text{H}_{10}\text{N}_4\text{S}$: N, 30.77. Found: 30.79 (by Dumas' method).

The production of this combination was very irregular. In some cases the yield was as low as 0.8 g., and it was our experience that no two experiments were productive of the same quantitative results although we operated as closely as possible under identical experimental conditions.

Examination of the Ammoniacal Filtrate.—This was evaporated to remove the ammonia and alcohol by heating at 100° when we obtained a light-colored solid residue which was amorphous in appearance. It

was insoluble in all the common organic solvents except alcohol and acetic acid. In these two solvents it was extremely soluble. By extraction with boiling water we were able to separate a crystalline substance which was moderately soluble in this solvent. After digestion with boiling water and cooling the solution, a cream-colored substance separated which was easily purified for analysis by crystallization from this solvent. This product was insoluble in both alkalis and acids, and when heated in a capillary tube melted with decomposition at about 210° . It contained sulfur, indicating the presence of the mercapto group, but did not respond to a test for chlorine. A nitrogen determination (Dumas) agreed with the calculated value for an inner anhydride of 2-ethylmercapto-4-aldehyde-

Calc. for $\text{C}_7\text{H}_7\text{N}_3\text{S}$: N, 25.45. Found: 25.30.

In other words, we are apparently dealing here with a new type of pyrimidine combination in which functions a pyrimidine and an isoazol or isopyrrol ring.

The amorphous residue, which remained behind after thorough leaching with boiling water, was pulverized and suspended in about 40 to 50 cc. of 20% hydrochloric acid. This was then heated on the steam bath when the material dissolved completely and as the solution evaporated ethylmercaptan was gradually evolved. After concentration to a small volume, an excess of sodium hydroxide solution was added to destroy the hydrochloric acid. This reagent at the same time caused the precipitation of a small amount of amorphous material which was separated by filtration. On carefully neutralizing this alkaline solution with hydrochloric acid a yellow substance was precipitated which by analysis was identified as cytosine aldehyde. It was purified by crystallization from hot water and melted at about 255° with decomposition. The substance was free from sulfur and chlorine and was soluble in both alkali and acid solutions. The pyrimidine was moderately soluble in water and practically insoluble in all the common organic solvents. The yield of this interesting pyrimidine combination usually amounted to about one g. The low yield is undoubtedly due to the fact that a large amount of material is destroyed by the action of the hydrochloric acid during the process of hydrolysis. In fact, we found that the pure aldehyde was completely destroyed if heated for a prolonged period with 20% hydrochloric acid. Analysis (Dumas):

Calc. for $\text{C}_8\text{H}_8\text{N}_3\text{O}_2$: N, 30.21. Found: 29.95.

The precursor of this new aldehyde is undoubtedly the corresponding 2-ethylmercapto compound, namely, 2-ethylmercapto-4-aldehyde-6-amino-

pyrimidine, but we did not succeed in isolating this combination,

NEW HAVEN, CONN.

[CONTRIBUTION FROM THE LABORATORIES OF THE ROCKEFELLER INSTITUTE FOR MEDICAL RESEARCH.]

SYNTHESES IN THE CINCHONA SERIES. I. THE SIMPLER CINCHONA ALKALOIDS AND THEIR DIHYDRO DERIVATIVES.

BY MICHAEL HEIDELBERGER AND WALTER A. JACOBS.

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Interest in the cinchona alkaloids as material for chemotherapeutic study has recently been revived by the remarkable specificity for the pneumococcus shown by ethylhydrocupreine.¹ The present paper is part of an investigation undertaken to test the possibilities for synthetic work in this field and deals with a number of the cinchona alkaloids, their reduction products, and certain synthetic homologs of the latter.

The cinchonine, cinchonidine, quinine, and quinidine used were purchased in the open market, and the constants obtained in the case of their monohydrochlorides are given below for comparison with the corresponding data for the other alkaloids considered.

The reduction of the four alkaloids to hydrocinchonine, hydrocinchonidine, hydroquinine, and hydroquinidine was very easily accomplished according to German patent 252,136, method 2, using palladium chloride in dilute sulfuric acid solution. Details of the method are given below for the case of hydroquinine. The properties of the hydrogenated alkaloids agreed with those recorded in the literature for the naturally occurring substances, and a direct comparison was made in the case of hydrocinchonine with a quantity of this alkaloid isolated as a by-product in the oxidation of commercial cinchonine (which contains hydrocinchonine) to the carboxylic acid cinchotenine.²

Considerable quantities of hydrocupreine were demanded in our synthetic work, and although this alkaloid is used in large amounts in the preparation of its ethers³ we were unable to find recorded any satisfactory method for obtaining this substance in fairly large quantities. Hesse⁴ heated hydroquinine in sealed tubes with hydrochloric acid (sp. gr. 1.125)

¹ Morgenroth and Levy, *Berl. klin. Wochschr.*, **48**, 1560, 1979 (1911).

² Skraup, *Ann.*, **197**, 376 (1879).

³ *D. R. P.* 254,712.

⁴ *Ann.*, **241**, 279 (1887).