l-Ribonic amide was prepared from ammonia and the lactone. The latter was prepared from arabonic acid by rearrangement on heating with pyridine, according to Fischer[1] and Piloty's directions. The amide was recrystallized twice from 90% alcohol and dried at 70° for several hours. M. p. 137–8°. 0.8450 g. of substance in 25 cc. aqueous solution rotated 1.11° to the left in a 2 dcm. tube; hence $[\alpha]_D^{20} = -16.4$. Weerman found m. p. 136–7° and $[\alpha]_D^{20} = -15.7$.

d-Mannosaccharic diamide was prepared according to Fischer's[2] directions by dissolving pure *d*-mannosaccharic dilactone in strong ammonia water, filtering off the crystalline precipitate and washing it with alcohol and ether. It was recrystallized once from water and dried at 70° for 14 hours. M. p. 188–189.5° with decomposition. 0.1765 g. of substance in 50 cc. aqueous solution rotated 0.36° to the left in a 4 dcm. tube; hence $[\alpha]_D^{20} = -24.4$. A second estimation gave —24.5. Fischer found m. p. 189°.

d-Saccharic diamide was prepared by the interaction of diethyl *d*-saccharic ester and ammonia, following Heintz's[3] directions. It was recrystallized twice from alcohol and dried at 100° for 5 hours. M. p. 172–3°. 0.8572 g. of substance in 50 cc. aqueous solution rotated 0.91° to the right in a 4 dcm. tube; hence $[\alpha]_D^{20} = +13.3°$.

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

RESEARCHES ON PROTEINS. VI. THE DESTRUCTIVE DISTILLATION OF FIBROIN.

[PRELIMINARY PAPER.]

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This preliminary report of an investigation, now in progress in this laboratory, is made in order to announce our intention of incorporating in our Researches on Proteins the study of the products of decomposition when silk fibroin is subjected to destructive distillation *in vacuo*. The research will not be completed for several months, but in the light of the recent work of Pictet and Cramer[4] on the destructive distillation of ovalbumin it seems necessary to make known at this time our activity in this same field of research.

Very little is known regarding the nature of the products of decomposition which are formed by the distillation of protein material. While much attention has been paid to the chemistry of coal distillation and also destructive distillation of wood and cellulose material, so far as the writers are aware, no important literature has been contributed dealing with the dry distillation of proteins outside of that bearing on the production of Dippel's oil by distillation of bones. The only publications available to us which contribute any data on this subject are those of

[1] *Ber.*, **24**, 4214 (1891).
[2] *Ibid.*, **24**, 539 (1891).
[3] *Pogg. Ann.*, **105**, 211 (1858).
[4] *Helv. Chim. Acte*, **2**, 188–195 (1919); *C. A.*, **10**, 1076 (1919).

Limpricht[1] and Williams[2] describing the distillation of horn and wool, respectively, in the presence of potash. In both cases the only organic combinations that were definitely identified among the products of distillation were carbon dioxide and several aliphatic bases, namely: ethyl, butyl and amyl amines. These combinations undoubtedly resulted as normal products of decomposition of the corresponding α-amino acids present in the protein molecule.

$$R.CH(NH_2)COOH \longrightarrow CO_2 + R.CH_2NH_2.$$

Later in 1885 Mills[3] subjected wool to dry distillation at ordinary pressure and identified as products of decomposition, hydrogen sulfide, water and ammonium carbonate.

Silk fibroin was selected for our investigation for three reasons. In the first place the protein is available in large quantities and can be obtained in a very pure condition. Secondly, it is one of the protein combinations which is free from sulfur, and thirdly, it is characterized by its amino acid composition. 60% of this protein molecule is composed of three amino acids, namely, glycocoll, alanine and tyrosine; these three acids constituting about 33%, 16% and 10%, respectively, of the fibroin molecule. Whether a careful study of the products formed by destructive distillation of proteins in general will contribute data throwing new light on the molecular construction of these substances remains to be determined, but it is true that fibroin is an excellent protein for experimentation on account of its unique composition.

Our method of experimentation has been to distil fibroin from an iron retort or pipe so constructed that the latter can conveniently be heated at a high temperature on an ordinary combustion furnace. Ordinary iron pipe of 5″ diameter has proven very practical for this operation. Having charged the retort with a known quantity of silk, it is then connected by the necessary cooling and absorption apparatus with a Cenco vacuum pump, which is sufficiently efficient to evacuate the system employed in our work. We first operated with a unit of 200 g. of fibroin (silk noils), but we finally increased this to 1600 g., which we found to be a very convenient and practical unit for laboratory operations. Working under these conditions, we have successfully distilled several kg. of silk fibroin, and the products of distillation are now being studied in order to determine the relative quantities formed, the compositions and the characteristic properties.

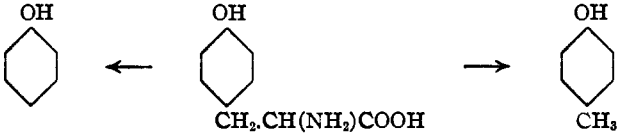
When distilled *in vacuo* (25–27 mm.) silk fibroin affords about 43% of its weight in the form of a red oil distillate, 41% in the form of silk carbon or coke and 16% in the form of volatile and gaseous products which

[1] *Ann. Pharm.*, **101**, 297 (1857).
[2] *Ibid.*, **109**, 127 (1858).
[3] *J. Soc. Chem. Ind.*, **4**, 325 (1885).

are absorbed by sodium hydroxide solution and sulfuric acid. The quantity of red oil distillate obtained varies from 660 to 790 g. per 1600 g. of fibroin, depending apparently to a large degree upon the pressure maintained during distillation of the protein. Thus far the only fraction which has received careful attention by us is the red oil distillate which is now under investigation. This is strongly ammoniacal and contains a large proportion of water besides organic substances of phenolic character.

We desire to report now on the isolation and identification of phenol among the components of this red oil distillate. This was separated in the following manner: An aliquot part of the oil distillate was subjected to steam distillation to remove the phenol and the latter then extracted from the steam distillate with ether and finally distilled under diminished pressure. We obtained an oil boiling from 48–58° under a pressure of 3 mm. This product was redistilled at ordinary pressure and finally washed with a 10% solution of sodium hydroxide to separate it from non-phenolic combinations. An oil having a pyridine odor was obtained here insoluble in the alkali. On acidifying the alkaline solution, after separating from insoluble oil, the phenol separated at once and was extracted with ether and again purified by distillation. It distilled as a colorless oil boiling at 182.5–185° at ordinary pressure. This product possessed all of the characteristic properties of carbolic acid, namely, odor and solubility in alkali and water, and it also gave the characteristic color test with ferric chloride. It reacted with bromine and nitric acid to give the characteristic insoluble tribromophenol, $C_6H_2(OH)Br_3$ (m. p. 89°), and picric acid, $C_6H_2(OH)(NO_2)_3$ (m. p. 122°), respectively.

Whether cresol is one of the products of distillation of fibroin has not yet been established. Both of these two phenols might result from the breaking down of tyrosine in the protein molecule. Quantitative experiments will be made in order to determine what portion of the tyrosine in



the protein molecule breaks down by distillation in this manner. It will also be of interest in this connection to subject polypeptide combinations of tyrosine to dry distillation *in vacuo*.

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