Von Lengyel's paper was received by the German Chemical Society on April 2nd, but the number of the *Berichte*[1] containing it only reached Washington on May 26.

Admitting that radio-active bodies can be manufactured to order, are we any nearer explaining their mysterious powers?

HENRY CARRINGTON BOLTON.

COSMOS CLUB, WASHINGTON, D. C., May 26, 1900.

MODERN RESEARCHES ON THE CHEMISTRY OF THE PROTEID MOLECULE.[2]

It is impossible, at the present stage of our knowledge, to give any satisfactory definition of a proteid, based either on its physiological or chemical properties. Physiologically, it can be pointed out as the main constituent of all cells and tissues. In regard to its chemical properties, it can be stated with absolute certainty that it consists of carbon, hydrogen, oxygen, nitrogen, and sulphur. It does not possess very marked acid or basic properties, but forms salts with both bases and acids, its affinity for both being very weak.

I.

It can not be classified under any of the well-established groups of chemical compounds. Some attempts in this direction, however, have been made in recent years; and of these, the attempt to classify all proteids among glucosides has been the cause of much dispute from the experimental and speculative side of the question. The author of this theory and its most enthusiastic advocate was Pavy, who, by hydrolysis of egg albumin, succeeded in obtaining a reducing substance, capable of combining with phenylhydrazine, forming an osozone of a definite melting-point.

Physiologists, who were all inclined to see the source of the tissue-carbohydrates in the tissue-proteids, naturally welcomed Pavy's work, and were ready to endorse his views. A number of researches, however, were undertaken in order to test the correctness of Pavy's statements. The results thereof were contradictory. Morner has investigated, in that direction, serum globulin, and found that on heating with 3–5 per cent. hydrochloric acid, it yielded a solution capable of reducing Fehling's solution. Krawkow has tested, in the same direction, various proteids with different results. Substances combining with phenylhydrazine, giving osozone, were obtained by him from

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[1] Vol. 33, p. 1237, May 14, 1900.

[2] Read before the New York Section of the American Chemical Society, May 11, 1900.

egg albumin (m. p. 183°–185° C.), fibrin (m. p. 182°–184° C.), and serum albumin (m. p. 183°–185° C.). He failed to obtain similar substances, or obtained them only in traces from serum globulin, lactalbumin, casein, gelatin, vitellin, and mucoid.

The work which followed that of Krawkow was done by Eichholtz and is instructive in many ways. Thus, in contradiction to Krawkow, he failed to obtain the carbohydrate from serum albumin and succeeded in obtaining it from serum globulin. Of greater interest, however, is the fact that he found in the white of the egg, besides the ovomucoid, a substance related to it, which he called "ovomucin."

In regard to the egg albumin, two other researches are of great importance: first, that of Weydemann, who obtained on treatment of egg albumin with a 10 per cent. solution of sodium hydroxide, a substance similar to "animal gum"; and secondly, the work of Spenzer, who repeated, under Drechsel's direction, the experiments of Pavy. Spenzer was very careful to remove all the mucoid from the white of the egg, and only used such methods as would exclude the contamination of the egg albumin with the carbohydrates of the filter-paper, etc. Under such conditions, Spenzer failed to obtain a carbohydrate on the hydrolysis of egg albumin with acids.

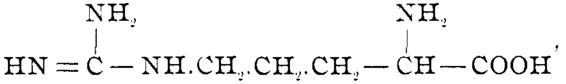
A comparison of the results of all the authors reveals the fact that proteids of the same nature gave different results to different investigators, as can be seen from the following table:

	Pavy.	Krawkow.	Eichholtz.	Morner.	Spenzer.
Egg albumin	positive	positive	positive	......	negative
Fibrin	"	"	......	......	......
Serum albumin	"	"	negative	......	......
Serum globulin	"	negative	positive	positive	......
Lactalbumin	......	"	......	......	......
Casein	positive	"	......	......	......
Gelatine	negative	"	......	......	......
Vitellin	positive	"	......	......	......
Mucoid	......	"	......	......	......

Further, new substances related to mucins were discovered where they were not suspected by some of the investigators searching for carbohydrate in the proteid molecule; and finally, those who were more careful in avoiding contamination were the least successful in obtaining the "carbohydrate moiety" of the proteid molecule. It seems therefore unwarranted at the present moment to accept the existence of such a moiety.

II. THE NITROGEN.

The greatest part of the researches on the chemistry of proteids has been directed to the character and form of the nitrogen present in its molecule. The older authors knew that the nitrogen was not all in equally strong combination with the rest of the proteid molecule, and that the nitrogen entered the proteid molecule in the form of an amido group. But of the amido compounds only those of the monamido acids were detected in that molecule. A great part of the nitrogen was in a combination unknown to most of the old investigators. Drechsel was the first to investigate with great success the nature of the nitrogenous residue. He has found that it consists of substances with a well-defined basic nature, some of them being diamido acids. The substances he discovered were lysin and lysatine: one being diamidovalerianic acid, $C_6H_{14}N_2O_2$; the other a homologue of creatine, $C_6H_{13}N_3O_2$. Later he also discovered among the decomposition products of proteids diamidoacetic acid. A number of different proteids were examined by the students of Drechsel, namely, E. Fisher, Siegfried, and Hedin, and the presence of the bases lysin and lysatine was demonstrated in all of them. Furthermore, Siegfried isolated besides these two bases, a new one having the composition $C_{11}H_{20}N_6O_6$. Hedin then obtained, by the decomposition of different proteids, arginin, a base of the following formula:



$$HN = \overset{\overset{NH_2}{|}}{C} - NH.CH_2.CH_2.CH_2 - \overset{\overset{NH_2}{|}}{CH} - COOH,$$

and which was first described by E. Schulze as a constituent of vegetating seeds. Later Hedin also demonstrated that the substance, described by Siegfried as $C_{11}H_{20}N_6O_6$, was histidin—$C_6H_9N_3O_2$—a base first discovered by Kossel as a decomposition product of a protamine, "sturin."

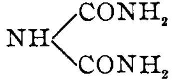
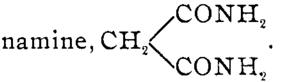
The statement of R. Cohn that a pyridin base could be detected among the other basic decomposition products of the proteids was very recently retracted by him.

After the presence of the basic substances in the proteid molecule was demonstrated, the question arose, how are they grouped in the molecule? and the researches of Kossel seemed to fully answer it. Kossel has resumed the work of Miescher on protamines—substances occurring mostly in fish sperm in combination with nucleic acid. The protamines had some properties common with proteids; namely, they gave the same color reaction with an alkaline copper solution as the proteids, a test known as the "Biuret test," undergoing also the same changes in solubility as proteids on digestion with pepsin hydrochloric acid.

Finally they yielded on decomposition the basic substances met with on decomposition of other proteids. The points of difference were that the protamines did not give the other color tests peculiar to proteid material and did not contain the other decomposition products met with on decomposition of proteids except the "hexon" bases. The conclusion was natural that the property of the proteids to give the biuret test was due to the presence in its molecule of a protamine group. Such actually was the conclusion of Kossel, and, according to his theory, the protamine is the nucleus of all proteids in the same manner as benzene is the nucleus of all the aromatic compounds.

On further investigation, however, it was found that only one protamine, namely sturin, yielded, on hydrolysis with acids, all the three "hexon" bases, sturin yielding as well amidovalerianic acid. The other protamines, such as clupein, scombrin, salamin, yielded only arginin, amidovalerianic acid, and an unknown residue. Cycloytelin contained besides these an aromatic group. Thus it appeared that the biuret color test is not peculiar to one certain "protamine group," and from this standpoint there is no reason to believe that all the proteids are derivatives of one protamine.

The analysis of animal proteids by Larrow and of plant proteids by Schulze, Mendel, and Levene, which have been made up to the present time, have revealed in all of them the presence of the three hexon bases. This would seem to corroborate the view that in proteids the biuret reaction is due to a "protamine." An objection to the latter assumption, however, can be found in the researches of H. Schiff.

H. Schiff has demonstrated that the biuret reaction can be obtained from different substances which contain two $CONH_2$ groups combined together, either directly, like oxamide  $\begin{array}{l}CONH_2\\ |\\ CONH_2\end{array}$ or joined on a single carbon or nitrogen, like biuret  $NH\begin{array}{l}\diagup CONH_2\\ \diagdown CONH_2\end{array}$ or malonamine,  $CH_2\begin{array}{l}\diagup CONH_2\\ \diagdown CONH_2\end{array}$. Another requisite is that the $CONH_2$ groups be combined in an open chain.

Thus H. Schiff has established the fact that the property giving the biuret color test is peculiar to more than one substance, that this property is due to the presence in the molecule, not of amido groups or amido acids, but of two of the radical $CONH_2$ grouped in a certain way; that it is not very probable that a combination of "hexon" bases (which, according to Kossel's first surmise, constituted the protamine molecule) will necessarily give the biuret test; that, finally, in the protamines as well as in other proteids the peculiar color formed on addition of alkaline copper solution is due to the presence in their respective molecules of a substance common to all of them, possibly, but perhaps not yet discovered. Attempts were also made to estimate the quantities of the different nitrogenous compounds in the molecules of different proteids, so as to establish a basis for their chemical classification. Hansemann has analyzed with this in view a great number of proteids, finding that the proportion of these constituents varied greatly with the character of the proteid. Henderson has, however, demonstrated that the temperature and the duration of the decomposition has a great influence on the formation of the different nitrogenous constituents, and thereby the conclusions of Hansemann lose much of their weight.

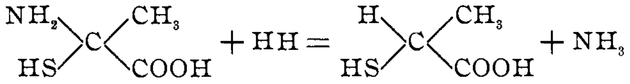
III. SULPHUR.

It has been accepted that the proteid molecule contains more than one atom of sulphur, and that the different atoms are in different forms of combination. This view is based on the fact that proteids heated with a solution of sodium hydroxide generally gave up part of their sulphur in the form of hydrogen sulphide, and the rest of the sulphur was detected only by means of strong oxidation, like fusion with sodium hydroxide and nitrate. Attempts have been made to establish the ratio between the different atoms. However, the methods employed by the older authors were not faultless, as the possibility of an oxidation of a part of the hydrogen sulphide into sulphuric acid (on heating with sodium hydroxide) was not excluded. The question thus needed new consideration, and such was given to it by F. N. Schultz. Great care was taken by the latter to prevent the possibility of oxidation of hydrogen sulphide, and in most experiments only about one-third of the total sulphur could be obtained in the form of hydrogen sulphide.

Very little has been known in regard to the nature of that part of the sulphur which could not be obtained as hydrogen sulphide. The opinion that it is in an oxidized state has been proved to be erroneous.

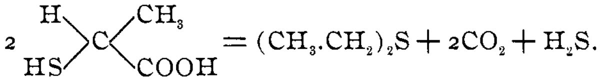
In recent years a few researches have appeared which throw some light on the subject, although none of them gives a final solution to the problem. Drechsel found among the basic decomposition products of the proteids, a substance which, on treatment with alkalies, yielded ethyl sulphide. He, therefore, concluded that the substance must be a sulphin base or a thetin compound; and that a quadrivalent sulphur was present in the proteid molecule.

A short time after Drechsel's discovery, Suter working in Baumann's laboratory, isolated from the decomposition products of proteids thiolactic acid, and very recently Morner has succeeded in obtaining, under the same conditions, cystein, which is a derivative of the former, as can be easily seen from the formulas.



$$\genfrac{}{}{0pt}{}{\mathrm{NH_2}}{\mathrm{HS}}\!>\!\mathrm{C}\!<\!\genfrac{}{}{0pt}{}{\mathrm{CH_3}}{\mathrm{COOH}} + \mathrm{HH} = \genfrac{}{}{0pt}{}{\mathrm{H}}{\mathrm{HS}}\!>\!\mathrm{C}\!<\!\genfrac{}{}{0pt}{}{\mathrm{CH_3}}{\mathrm{COOH}} + \mathrm{NH_3}$$

Baumann has demonstrated further that similarly to the thetin compounds, the cystein or thiolactic acid yields ethyl sulphide on treatment with alkalies, according to the following formula:



$$2\,\genfrac{}{}{0pt}{}{\mathrm{H}}{\mathrm{HS}}\!>\!\mathrm{C}\!<\!\genfrac{}{}{0pt}{}{\mathrm{CH_3}}{\mathrm{COOH}} = (\mathrm{CH_3.CH_2})_2\mathrm{S} + 2\mathrm{CO_2} + \mathrm{H_2S}.$$

Thus, another explanation was offered for the appearance of ethyl sulphide among the decomposition products of proteids. It must be remembered that both the substances ethyl sulphide and cystein (cystin) had been detected in animal secretions long before they could be obtained directly from proteids. The former was first identified in the urine of a dog by J. Abel; the latter by Baumann.

HALOGEN PROTEIDS.

Considerable study has been devoted to the halogen derivatives of the proteids; it was expected that the latter compounds would be of great aid in explanation of the constitution of the proteid molecule. The expectations, however, have not been realized up to the present date; although the future may be more successful.

SYNTHESIS.

Notwithstanding that little regarding the chemical constitution of the proteid molecule is established, attempts have already been made to obtain a proteid synthetically. In recent years the attempt was made by Lilienfeld, who stated in 1894 that he was successful in obtaining a synthetical proteid in the following way:

Curtius and Goebel found that if glycocoll-ethyl ester was allowed to stand, it yielded glycocoll and a biuret-giving substance which Lilienfeld claimed could form a condensation product with the ethyl ester of leucin, tyrosin, or aspartic acid, which product resembled peptone very closely.

More recently, Lilienfeld has modified his method and obtained proteids on condensation of phenol with glycocoll or with asparagin, etc.

Klimmer, however, justly remarks that Lilienfeld's substance could scarcely be considered a peptone on the following grounds:

1. The substance obtained by Lilienfeld yields readily, on decomposition, phenol and glycocol which other proteids do not, and the color which the substance takes, on treatment with alkaline copper solution, is not a color resembling the biuret reaction very closely.

Thus, little progress has been made towards the elucidation of the chemistry of the proteid molecule and its various parts. Attempts have been made to determine the weight of the molecule as a whole. According to Sabanejeff and Alexandrow, it is 14,900.

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