the late Henry van Peters Wilson, the business meeting adjourned.

At 8:30 P.M. the evening meeting was held with the vice-president, Donald B. Anderson, presiding. After a brief address of welcome by Dean Bryan, of Wake Forest College, President John W. Lasley, Jr., gave his retiring address entitled "The Relation between Mathematics and the Sciences."

On Saturday afternoon the academy met in the following sections: General, Botany, Mathematics, Physics, Zoology, The North Carolina Section of the American Chemical Society and the High School Science Teachers.

The following officers were elected by the various sections:

ZOOLOGY SECTION

Chairman: J. P. Givler, Woman's College, University of North Carolina.
Secretary: Z. P. Metcalf, North Carolina State College.

MATHEMATICS SECTION

Chairman: T. F. Hickerson, University of North Carolina.
Secretary: J. A. Greenwood, Duke University.

PHYSICS SECTION

Chairman: H. E. Fulcher, Davidson College.
Secretary: F. W. Lancaster, North Carolina State College.

NORTH CAROLINA SECTION OF THE AMERICAN CHEMICAL SOCIETY

Chairman: E. C. Markham, University of North Carolina.
Vice-Chairman: W. E. Jordan, North Carolina State College.
Secretary-Treasurer: Ivan D. Jones, North Carolina State College.

HIGH SCHOOL SCIENCE TEACHERS

Chairman: Clifford Beck, Salisbury, N. C.
Secretary: Harry MacDonald, New Bern, N. C.

H. L. BLOMQUIST,
Secretary

SPECIAL ARTICLES

CHEMICAL COMPOSITION OF THE TUMOR-PRODUCING FRACTION OF CHICKEN TUMOR I

THE agent transmitting chicken tumor I can be separated from tumor extracts quantitatively, and partially purified, by a method of centrifugation at high speed.[1] The material prepared in this manner is found to be composed of small granules, visible in the dark-field microscope, approximately 70 mμ in diameter,[2] with a corresponding particle weight of about 2.3×10^{-16} grams and a molecular weight of *ca.* 140 millions.[3] The tumor-producing power of ordinary tumor extracts varies a great deal, and only preparations exhibiting high activity were retained for this study. Tests were made by injecting the freshly prepared material intracutaneously in adult hens. Tumors were usually present after 8 days and the final results were recorded not later than 18 days after injection. In these experiments, the smallest dose to produce actively growing tumors was about 4×10^{-13} grams, in terms of dry weight, of the purified substance. The relation between this weight and the weight of the individual particles indicates that the minimal active dose contained approximately 2,000 of the elementary granules.

The purified tumor fraction contains 8.5 to 9.0 per cent. nitrogen, a confirmation of previous analysis, and 1.5 per cent. phosphorus. Tests for proteins are positive. A typical Feulgen reaction, characteristic of thymonucleic acid, is absent. However, an intense reaction is obtained with the Schiff reagent, which suggests the presence of aldehyde groups in the material. Tests for pentoses are strongly positive.

Previous work has shown that the purified tumor fraction could be decomposed into two chief components, namely, a lipoid fraction and a protein fraction.[4] The chemical and physical properties of these fractions have been investigated further, and the results of this study is the matter of the present note.

The lipoid material associated with the tumor-producing fraction of chicken tumor I, and extracted by ether, had been found to represent about 24 to 26 per cent. of the whole substance.[4] The results of a somewhat more vigorous extraction by treating the dry substance with ether-alcohol and chloroform, at room temperature, indicates that at least 35 per cent. of the total dry weight of the purified material is lipoid in nature. This complex lipoid material contains about 2.4 per cent. phosphorus and 1.6 per cent. nitrogen.

Two main fractions can be separated from it. (*a*) One fraction is soluble in cold acetone. At room temperature it is a soft, yellow-colored mass which crystallizes in narrow plates arranged in rosettes. This fraction contains about 1.8 per cent. phosphorus and 0.9 per cent. nitrogen, figures which suggest that about half of the material may be in the form of phospholipids. The test with the Schiff reagent is negative or weakly positive. (*b*) The second fraction, which is the most abundant, has the general composition and

[1] J. C. G. Ledingham and W. E. Gye, *Lancet*, 1: 376, 1935; C. R. Amies, *Jour. Path. Bact.*, 44: 141, 1937; A. Claude, *Jour. Exp. Med.*, 66: 59, 1937, *Am. Jour. Cancer*, 30: 742, 1937, and SCIENCE, 87: 467, 1938.

[2] W. J. Elford and C. H. Andrewes, *Brit. Jour. Exp. Path.*, 17: 422, 1936.

[3] A. Claude, *Jour. Exp. Med.*, 66: 59, 1937; K. G. Stern and F. Duran-Reynals, SCIENCE, 89: 609, 1939.

[4] A. Claude, *Jour. Exp. Med.*, 61: 41, 1935; SCIENCE, 87: 467, 1938.

physical properties of phosphatides. It is precipitated by cold acetone and contains 3.8 per cent. phosphorus and 1.8 per cent. nitrogen, 0.3 per cent. of which is in the form of amino-nitrogen. A characteristic property of this lipoid component is that it gives a strongly positive reaction for aldehydes, when tested with the fuchsin-sulfurous acid solution of Schiff. The reaction is accelerated by acid, by heat and acid, by mercury bichloride and by trichloracetic acid. This aldehyde component of the tumor material resembles closely the substance demonstrated in the cytoplasm of certain cells, under the name of plasmalogen, by Feulgen and Voit.[5] The plasmal reaction of Feulgen, carried out on frozen sections, showed the cytoplasm of chicken tumor cells to be loaded with this lipoid.

The occurrence, in the purified chicken tumor material, of relatively large quantities of a substance which may exhibit the properties of an aldehyde is of interest, as regards tumor production, but this point is still undetermined. It is also possible that this chemically active group, which can be uncovered easily, plays a part in the so-called spontaneous inactivation of the tumor agent.

The complete lipoid fraction can be taken up in water or in salt solution, making a stable and homogeneous colloidal suspension. This is accomplished more easily under moderate heat or when traces of alcohol, which can be removed later, are present. Under the dark-field microscope this suspension appears to be composed of small particles which, as regards size and shape, are indistinguishable from those making up the original, unfractionated material. This observation suggests that the lipoid components of the purified tumor fraction may play an important role in the morphology of the active granules.

The portion of the tumor material, which is left after lipoid extraction, contains 12.6 to 13.2 per cent. nitrogen and about 1 per cent. phosphorus. Tests for protein are positive. In contrast to the complete fraction, the test with the Schiff reagent is negative, in agreement with the fact that the substances giving the reaction are removed by organic solvents. Thymonucleic acid is absent, or is not present in appreciable quantities, as shown by negative Feulgen and Dische tests. Tests for pentoses are positive. These observations support the view, stated previously, that the nucleoprotein which forms a large portion of the purified chicken tumor fraction is of the ribose type.[6]

A nucleic acid has been prepared from this material, by alkaline hydrolysis.[7] The separation is readily accomplished by leaving the freshly prepared tumor suspension in 0.1 N NaOH for 6 to 12 hours at 4° C. The alkaline mixture is then brought to pH 4.7 with 0.1 N acetic acid. The flocculent precipitate which settles at this point is removed by the centrifuge. The supernatant liquid is clear and colorless, and gives a negative biuret reaction. The Feulgen test for thymonucleic acid is negative, but tests for pentoses are strongly positive. The solution absorbs ultra-violet light in the manner of nucleic acid, with the position of the maximum at λ2575 and the minimum near λ2400.[8] The intensity of absorption of ultra-violet light and the phosphorus content suggests that 10 to 15 per cent. of the protein may be nucleic acid.

From the above nucleic acid fraction, guanylic acid was isolated by further alkaline hydrolysis. The substance gave positive tests for pentoses and the characteristic absorption spectrum of guanine.[9]

Previous studies have shown that active fractions of chicken tumor I, prepared by various methods, present a characteristic ultra-violet absorption spectrum, with a broad maximum in the region of λ2550–2575.[10] The present work is in agreement with these findings, as both the complete tumor fraction and the "nucleic acid" derived from it also show the typical maximum of absorption at $\lambda \simeq 2575$.[8] These observations indicate, either that the substance responsible for the absorption in that region is not yeast nucleic acid, which has its maximum at λ2600,[10] or that the material contains an absorbing element which causes the maximum to be consistently displaced toward the shorter wave-lengths. The separation of a guanine nucleotide which has a broad absorption band in the region of $\lambda = 2500$, and evidence from unpublished experiments,[11] suggests that this effect might be caused by guanine as a predominating element in the molecule. A quantitative study of the various nucleotides must await the preparation of suitable quantities of material.

By the same method of differential centrifugation at high speed, it is possible to separate from normal chick embryo a fraction which, in many respects, resembles the active fraction of chicken tumor I.[12] Further analysis of this chick embryo material has emphasized the similarities between the two substances. The chick embryo material, like the tumor fraction, is found to consist essentially of a phospholipid-aldehyde portion, associated with a nucleoprotein of the ribose type.[13, 14]

ALBERT CLAUDE

ROCKEFELLER INSTITUTE,
NEW YORK, N. Y.

[5] R. Feulgen and K. Voit, *Arch. Ges. Physiol.* (*Pflügers*) 206: 389, 1924.

[6] A. Claude, SCIENCE, 87: 467, 1938.

[7] P. A. Levene and L. W. Bass, "Nucleic Acids," The Chemical Catalog Co., Inc., New York, 1931.

[8] The ultra-violet absorbing power of the solutions was determined by Dr. A. Rothen.

[9] F. F. Heyroth and J. R. Loufbourow, *Jour. Am. Chem. Soc.*, 56: 1728, 1934; T. Caspersson, *Scand. Arch. Physiol.*, 73: Supplement No. 8, 1936.

[10] A. Claude and A. Rothen, *Am. Jour. Cancer*, 26: 344, 1936; A. Claude, *Am. Jour. Cancer*, 30: 742, 1937, and SCIENCE, 87: 467, 1938.

[11] In collaboration with Dr. G. I. Lavin.

[12] A. Claude, *Proc. Soc. Exp. Biol. Med.*, 39: 398, 1938.