be quantitatively expressed by suitable application of the laws of classical chemistry.[2]

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THE AMOUNT OF CIRCULATING PRECIPITIN FOLLOWING THE INJECTION OF A SOLUBLE ANTIGEN

IN the preceding note preliminary data were reported on the precipitin reaction between a protein and its homologous antibody. These data permit for the first time a calculation of some theoretical interest; namely, the amount (weight as opposed to titer) of circulating precipitin in an animal following immunization by a given amount of antigen. For example, rabbits 49 and 54 were injected with small doses of *R-salt-azo-benzidine-azo-egg albumin* until a total of 21.6 mg of the dye had been given. The animals were bled 10 days after the last injection, and antibody solutions were prepared by sodium sulfate fractionation and made up to double the serum volume. This may be considered as blood volume for the present purpose. The maximum specifically precipitable protein[1] in solution 49 was 1.53 mg per cc; in solution 54, 1.25 mg per cc. Taking the weight of the rabbits as 2 kg and their blood volume as 5.5 per cent. of their weight,[2] or 110 cc, the blood of rabbit 49 contained 168 mg of precipitin at the time of bleeding, while that of rabbit 54 contained 138 mg. Calculated as milligrams of circulating precipitin per milligram of antigen injected, the values are 7.8 and 6.4, respectively.

Naturally these figures are inaccurate, since the exact blood volumes of the rabbits were not known. From the theoretical standpoint, however, it is of interest that they are probably low, since losses undoubtedly occurred in the preparation of the antibody solutions. Moreover, these values can only represent a fraction of the total antibody formed, since storage in the cells occurs as a result of sensitization of tissues and organs. It is also not certain that the circulating precipitin is the only circulating antibody. On the other hand it is considered, as in the preceding studies, that antibody is modified globulin, and that the antibody precipitated is not contaminated with non-specific serum globulins. Evidence on the latter point will be reported later.

According to Svedberg and Sjögren[3] the molecular weight of serum globulin is three times that of egg albumin. If one assumes antibodies to have about the same molecular weight as the globulins with which they are associated, and the egg albumin dye to have about the same molecular weight as egg albumin, each dye molecule would have to split into more than two specifically reactive fragments if it participated in the building up of the antibody molecule. However, Landsteiner has repeatedly shown that the specificity of the azo protein dyes is a function of the dye component, rather than of the protein used. It would therefore be reasonable to expect that if the antigen or any of its fragments participated in the building up of the antibody molecule the antibody would be colored. It is true that the crude antibody solutions obtained by fractionation of the sera of animals immunized to the red protein dye were definitely pink, but the color disappeared almost completely on dialysis.

The preliminary data herein presented therefore tend to favor the view that the antigen itself does not participate in the building up of the antibody complex. Further information on this and related questions is being sought along these lines and it is hoped that more decisive figures will be obtained.[4]

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CORRELATION OF ANTILLEAN FOSSIL FLORAS

CERTAIN Antillean plant beds have been described by Drs. Hollick,[1] Vaughan and Berry,[2] Hodge,[3] Howe[4] and Maury,[5] and a tentative grouping and correlation is now suggested.

The Nilssonia bed, Porto Rico.—South of Cidra, Mr. Hodge found plants in a bog iron ore bed.

2 This study was carried out under the Harkness Research Fund of the Presbyterian Hospital.

1 *Jour. Exp. Med.*, 50: 809, 1929.

2 Meek and Gasser, *Am. Jour. Physiol.*, 47: 302, 1918–19.

3 *Journ. Am. Chem. Soc.*, 50: 3318, 1928; 52: 2855, 1930.

4 This study was carried out under the Harkness Research Fund of the Presbyterian Hospital.

1 "Rio Collazo Plant Beds, Porto Rico," "Scientific Survey of Porto Rico and the Virgin Islands," Vol. 7, pt. 3, 1928; "Siparia Flora, Trinidad," *Bull. N. Y. Bot. Garden*, vol. 12, No. 45, 1924; "Rio Guajataca Flora, Porto Rico," *Jour. N. Y. Bot. Garden*, 27: 223–7, 1926.

2 "Sánchez Flora, Dominican Republic," "Geological Reconnaissance of the Dominican Republic," p. 165, 1921.

3 "Algae of Coamo Springs Limestone, Porto Rico," "Scientific Survey of Porto Rico," vol. 1, pt. 2, pp. 153–9, figs. 15, 16 (not 18), pp. 195, 228, 1920.

4 "Algal Flora, St. Bartholomew, Antigua, Anguilla," Carneg. Inst. Wash., Pub. No. 291, pp. 11–19, 6 plates, 1919.

5 "Los Quemados Flora, Dominican Republic," *Bull. Amer. Paleontology*, No. 30, p. 19, 1917.

These were identified by Drs. Berry and Knowlton as *Nilssonia*, an old Mesozoic cycad genus; a species of the fern *Protorhipis*, and a dicotyledon. Drs. Britton and Hollick, in 1926, 1927 and 1928, examined this locality but found only unidentifiable plant fragments.

The *Nilssonia* flora lay beneath a horizon with corals referred by Mr. Hodge to *Cladophyllia furcifera*, suggesting an age corresponding to the Fredericksburg group of the Lower Cretaceous of the gulf states. But the foraminiferal evidence of the Rio de la Plata series said to underlie the Baranquitas-Cayey series (of which the *Nilssonia* bed is a member), as identified by Dr. Bagg, suggests an Upper Cretaceous age. This contradiction requires clearing up. Meantime the possibility of a Fredericksburgian age for the *Nilssonia* bed is very intriguing because of the practical absence of fossils of Lower Cretaceous age in the Dominican Republic and Haiti, and elsewhere than Cidra in Porto Rico, and even there the evidence is not certainly proved. Further studies may prove the *Nilssonia* bed to be Upper Cretaceous.

The Archaeolithothamnium Antillean beds.—Fossil calcareous algae are abundant in certain deposits on St. Bartholomew, Antigua, Anguilla and Porto Rico. *Archaeolithothamnium affine* Howe was described by Dr. Howe from the Antiguan Oligocene, and was compared to *A. turonicum* from the Turonian stage of the Upper Cretaceous of France. The *Archaeolithothamnium* found by Mr. Hodge in the Coamo Springs algal limestone, Porto Rico, shows affinities with both the Oligocene *A. affine,* and with the Upper Cretaceous *A. turonicum,* hence its stratigraphic position can not be set by this alga alone. Dr. Coryell has reported *Archaeolithothamnium* also in limestones on the Coamo-Aibonito road, Porto Rico.

Flora of the Rio Collazo shales, Porto Rico.—This flora was found by Dr. Hollick to be very rich in dicotyledons, with palms abundant, but only two cycads were present, one fern and one alga. Dr. Hollick noted its typical New World facies, tropical character and Tertiary age.

From the molluscan evidence I refer the Rio Collazo flora to the Antiguan Oligocene.

The Siparia flora, Trinidad.—This flora was described by Dr. Hollick in 1924, but no age assigned. My own paleontological studies in Trinidad lead me to regard the Siparia flora as of Miocene age. But its exact position in that period is not determined.

The Los Quemados flora, Dominican Republic.—In 1916, the Maury Expedition collected *Eugenia, Nectandra, Inga* and various species of woods from the Gurabo formation on Rio Gurabo, near Los Quemados. This flora was shown by molluscan evidence to be of Middle Miocene age. An arm of the sea with rich molluscan and coral life occupied the present valley of Rio Yaque del Norte, while the neighboring shores were bordered with myrtles, laurels, mimosas and a variety of forest trees.

The Sánchez flora, Dominican Republic.—In 1921, the Vaughan Expedition obtained plants from the clays near Sánchez in the northeastern part of the Dominican Republic. This flora apparently is later than that of Los Quemados, but its exact horizon has not yet been determined. Dr. Vaughan referred it to Miocene or Pliocene.

The Rio Guajataca flora, Porto Rico.—A Post-Pliocene walnut was described by Dr. Hollick from the valley of Rio Guajataca, in 1926, and named *Juglans archaeoantillana.*

Antillean climates of the past.—Antillean fossil floras indicate that the West Indian climate has remained unchanged from Antiguan Oligocene time to the present day.

Antillean sea shells in Tertiary time were also tropical in character, for their living allies are inhabitants of warm seas. In a very few instances, however, competition has forced their modern descendants to the deep sea where they live in almost freezing water. For example, the modern analogue of *Crepitacella melanoides* Gabb of the Dominican Middle Miocene is *C. gabbi* Dall, living in ooze at 785 fathoms. With rare exceptions, the shells like the plants indicate that the temperature of the sea and of the air was similar to that of to-day in the Antilles.

In connection with Antillean stratigraphy, in reply to Professor Meyerhoff's criticism[6] of my recent paper,[7] I would say that my statement was based on the fact recorded by Dr. Vaughan[8] that R. T. Hill collected Caprinidae *in limestones* near Fajardo, eastern Porto Rico, and at Cape San Juan which forms the northeastern extremity of the island. I would also like to add that the identification of *Venericardia alticosta* Conrad, from the Rio Jueyes Water Gap, Porto Rico, was made by Professor Grabau, and hence is highly authoritative.

A further note on the age of the Porto Rican Sirenian may be of interest to vertebrate paleontologists. In 1915, Dr. Reeds found a fossil Manatee in the shales at the ford on Jacaguas River, one kilometer northeast of Juana Diaz, southern Porto Rico. This was named *Halitherium antillense* by Dr. Matthew, in 1916, but its age remained indeterminate. The invertebrate fauna of the Juana Diaz shales proves the Manatee to have lived in Antiguan Oligocene time.

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[6] SCIENCE, 71: 322–3, March 21, 1930.
[7] SCIENCE, 70: 609, December 20, 1929.
[8] *Jour. Washington Acad. Science,* vol. 13, No. 14, 1923.