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

MICHAEL HEIDELBERGER
FORREST E. KENDALL

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 nonspecific 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]

MICHAEL HEIDELBERGER
FORREST E. KENDALL

DEPARTMENT OF PRACTICE OF MEDICINE,
PRESBYTERIAN HOSPITAL AND
COLLEGE OF PHYSICIANS AND SURGEONS,
COLUMBIA UNIVERSITY, NEW YORK

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.