

10046 P

Complement Fixation with Insulin as Antigen.

RUSSELL RICHARDSON. (With technical assistance of Peggy C. Kostal.)

From the George S. Cox Medical Research Institute, University of Pennsylvania, Philadelphia.

In testing a hypothesis regarding insulin it was observed that certain human sera specifically fixed complement in the presence of the regular insulin as supplied to patient, in U80 strength; as well as pork, beef, and crystalline insulin.*

To determine the optimal amount of insulin to be used in the test a titration was set up according to the method of Boerner and Lukens¹ following that of Hooker² for the Wassermann antigen, using a serum which had previously shown marked fixation of complement with insulin. The result (Table I) is similar to that

TABLE I.
Titration of Insulin as Antigen with a Strongly Positive Serum.

Serum (cc)	Units of insulin used as antigen				
	25U	20U	15U	10U	5U
0.0063	—	+	+	—	—
0.0125	—	++	++	+	—
0.025	++	++	++	++	++
0.05	++	++	+++	+++	+++
0.1	++	++++	++++	++++	++++
0.2	++	++	++	+	+

found when the Wassermann antigen is titrated. With too large or too small an amount of either antigen or serum the maximal fixation is not obtained.

When this optimal dose of insulin had been determined human sera were tested, in the following quantities—0.1, 0.05, 0.025, 0.0125, and 0.0063 cc; the usual controls were employed. Sera that gave complete or almost complete inhibition of hemolysis in one or more tubes were considered to be positive. A few sera were found in which there was a slight but definite inhibition of hemolysis, recorded as a doubtful reaction. Tests were repeated on the

* We wish to thank the Eli Lilly Company for the very generous supply of insulin furnished for this work.

¹ Boerner, F., and Lukens, M., *Am. J. Clin. Path.*, 1937, **7**, 33, 155.

² Hooker, S. B., *J. Immunol.*, 1927, **14**, 129.

original serum and from 1 to 8 times on sera collected at intervals of from 1 week to 2 months. In all except 5 persons these reactions showed results similar to those found in the first test. The variations were of the same order as are commonly found when the Wassermann test is repeated on a given serum. The results are shown in Table II.

TABLE II.
Complement-fixation Reaction with Insulin as Antigen.

	Positive	Doubtful	Negative	Total
Thyroid	4	2	5	11
Pregnancy	9	4	20	33
Diabetes	37	10	153	200
Other diseases	3	1	44	48
Normals	6	3	58	67

The normals in this table consisted of doctors, nurses, and members of the laboratory staff. They were apparently in good health but had not all had physical examinations. All were free from clinical signs of syphilis and 42 had negative blood-Wassermann reactions.

The thyroid-patients had been selected at random from the Thyroid Clinic and varied both in clinical condition and in the height of the basal metabolic rate.

The stage of pregnancy varied from the 3d to the 8th month and appeared to show no correlation with the reaction.

The diabetic patients were unselected. The group included balanced and unbalanced patients as well as those who were on diet alone and on diet with insulin. In all of these patients the Wassermann reaction was negative. No definite correlation was found between the clinical condition and the reaction.

The other diseases consisted of groups of from 5 to 10 patients each with a diagnosis of carcinoma, tuberculosis, allergic condition, acute inflammation, chronic inflammation, some kind of blood disease, or syphilis with a positive blood-Wassermann reaction. Seven sera giving positive Wassermann reactions were negative with insulin; the Wassermann was negative in 52 insulin-positive sera tested for that reaction. In no serum were the Wassermann and insulin reactions both positive. Two insulin-positive sera did not react with insulin that had been inactivated by treatment with alkali.

It would then appear that there is present in the blood of certain persons a substance that is able to combine with insulin and thereby fix complement. Its nature and the degree of specificity of its action with insulin are not yet defined.