which contains no sulfonamide. If the number of colonies is not within these limits the test must be repeated with a more suitable inoculum.

The tubes are incubated at 37°C for 18 to 24 hours; and the test is read in terms of the concentration of sodium sulfadiazine in the highest tube in which visible growth occurs. If this be in the control tube only the result is "0"; if in all tubes except that containing 125 mg of the drug per 100 cc of medium the result is "25." Tests showing values of 0 and 25 are shown in Fig. 1.

Discussion. Results have been reproducible with a variation of plus or minus one tube. Strains have been kept on blood agar slants in the ice box for at least one month without undergoing a change in their *in vitro* resistance to sulfadiazine.

To obtain consistent results the same sulfonamide and serum from the same species must be used. Sodium sulfathiazole has given readings about one tube lower than sodium sulfadiazine; and medium made with rabbit serum has given readings about one tube higher than that made with horse serum and xanthine.

The semi-synthetic part of the medium is doubtless unnecessarily complex, because several of its constituents are contributed in sufficient quantity by the added serum. No attempt has, as yet, been made to eliminate these superfluous materials or to reduce the medium to its simplest form.

Many strains of hemolytic streptococci are inhibited by as little as 0.2 mg of sodium sulfadiazine per 100 cc of the medium, which indicates that the content of substances antagonistic to sulfonamide action is very low.

No success has attended attempts to refine the test in such a manner that lesser degrees of sulfonamide resistance might be demonstrated. The intervals between the concentrations of drug in the tubes (multiples of 5) have been chosen deliberately; and the good correlation which exists between the findings of the test and clinical observations in the field[3] suggests that it is sufficiently precise to be of practical value.

Summary. A technic for testing the resistance of Group A hemolytic streptococci to sulfonamides is presented. The test has been found to be sufficiently precise to indicate clinically significant differences in sulfonamide susceptibility.

[3] Wilson, A. T., and Coburn, A. F., to be published.

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Competition of Antigens in Isoimmunization by Pregnancy.*

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When an experimental animal is injected with a mixture of two substances, one a good antigen and the other a poor antigen, the antigenicity of the weaker antigen is often suppressed.[1] The purpose of the present communication is to report some observations in man of this phenomenon, known as competition of antigens.

The properties A and B are good antigens in man, as revealed by observations made following inadvertent transfusions of blood of an incompatible group.[2] On the other hand, only 1 in 25 to 50 Rh-negative individuals respond to transfusions of Rh-positive blood, or pregnancy with an Rh-positive fetus, by

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[1] *Cf.* Landsteiner, K., *The Specificity of Serological Reactions*, 2d edition, p. 104, Harvard Univ. Press, Boston, 1945.

[2] Wiener, A. S., *Blood Groups and Transfusions*, p. 123, 3rd ed., C. C. Thomas, Springfield, Ill., 1943.

producing Rh antibodies.[3,4,5] Therefore, for example, if a group O Rh-negative woman bearing a group A Rh-positive fetus becomes isoimmunized, it would be expected that she would either produce antibodies for A alone, or antibodies for A and Rh simultaneously, but not for Rh alone. In general, moreover, it would be expected that an Rh-negative woman is more apt to become sensitized to the Rh factor present in her fetus if the fetus belongs to a compatible blood group than if the fetus' group is incompatible. That this is actually the case can be seen from Table I, in which the incidence of compatible matings in families with erythroblastotic infants is compared with the incidence of compatible matings in a random series of families without children having hemolytic diseases. (The excess of compatible matings in families with erythroblastotic infants has also been noticed by Levine,[6] but no attempt was made to explain it.) In cases in which an Rh-negative woman had an erythroblastotic baby, who belonged to an incompatible blood group, and titration experiments were carried out, the present author has usually found, in accordance with expectation, a high titer of the corresponding specific isoagglutinin in the maternal serum.

These observations can be applied in cases of stillbirths or neonatal deaths of obscure etiology, in which the mother is Rh-negative, the father and fetus (or infant) Rh-positive, but the maternal serum does not contain any detectable Rh agglutinating or blocking antibodies.[5] In the absence of such isoantibodies, the possibility still remains open that the mother may not be sensitive to the Rh factor and the fact that she is Rh-negative may merely be a coincidence. This interpretation is supported if the fetus (or infant) belongs to an incompatible blood group, and the corresponding isoagglutinin in the maternal serum is nevertheless of only average titer. On the contrary, the diagnosis of erythroblastosis is rendered more probable if the corresponding

TABLE I.
Comparison of the Incidence of Compatible Matings in Families with Erythroblastotic Infants, and Their Incidence in Normal Families.

Families with erythroblastotic infants				Control, normal families				
Investigators	Total No. of families	Compatible % ± P.E.	Incompatible %	Investigators	Total No. of families	Compatible % ± P.E.	Incompatible %	Difference % ± P.E.
Present study	96	71.9 ± 3.0	28.1	Wiener *et al.*§	215	63.2 ± 2.2	36.8	+ 8.7 ± 3.7
Levine*	215	75 ± 1.9	25	Calculated from distribution of groups in N. Y. C.	—	65	35	+10 ± 1.9
Race, Taylor, Cappell and McFarlane†	40	70.0 ± 3.5	30.0	Taylor, Race *et al.*¶	236	64.4 ± 3.1	35.6	+ 5.6 ± 4.7
Broman‡	33	81.8 ± 4.5	18.2	Calculated from distribution of groups in Sweden	—	64.2	35.8	+17.6 ± 4.5

* *J. Hered.*, 1943, **34**, 71.
† *Brit. Med. J.*, 1943, **2**, 289.
‡ *Acta Paed.*, 1944, Vol. XXXII, Suppl. II.
§ PROC. SOC. EXP. BIOL. AND MED., 1943, **53**, 167; *Genetics*, 1943, **28**, 157; *J. Exp. Med.*, 1944, **79**, 235.
¶ *Ann. Eug.*, 1938, **8**, 343; *ibid.*, 1942, **11**, 385.

[3] Wiener, A. S., and Peters, H. R., *Ann. Int. Med.*, 1940, **13**, 2306.

[4] Wiener, A. S., *Arch. Path.*, 1941, **32**, 227.

[5] Wiener, A. S., Wexler, I. B., and Gamrin, E. L., *Am. J. Dis. Child.*, 1944, **68**, 317.

[6] Levine, P., *J. Hered.*, 1943, **34**, 71.

specific isoagglutinin in the maternal serum is of high titer or if the fetus (or infant) belongs to a compatible blood group. Determining the titer of the maternal isoagglutinins is also of value in the less common cases of suspected hemolytic disease in which the mother is Rh-positive, because here one must still consider the possibility of isoimmunization against factor Hr, or the isoimmunization of an individual of one Rh type against blood of a different Rh type.

The phenomenon of competition of antigens also manifests itself with regard to the different varieties of Rh factors.[7] Of the 3 Rh factors, Rh_o is by far the most antigenic, Rh′ is less antigenic, and Rh″ is the least antigenic.[8] Therefore, it would be expected that if, for example, an Rh-negative woman bears a type Rh_1 fetus and becomes sensitized to the Rh factor, her serum should either contain anti-Rh_o alone, or anti-Rh_o and anti-Rh′ together, but not anti-Rh′ alone. Actual observations fully satisfy these expectations; the cases where the maternal sera appeared to contain anti-Rh′ alone all proved actually to be examples of anti-Rh'_o sera with anti-Rh_c blocking antibodies.[9,10,11] It is highly significant that even though a high percentage of erythroblastotic infants from Rh-negative mothers belong to type Rh_2, only two instances of anti-Rh''_o sera from such mothers have been encountered[12] and none of anti-Rh″ alone, in contrast to the numerous anti-Rh_o sera such cases have yielded. On the other hand, in the far rarer instances of the type Rh_1 mother bearing an erythroblastotic infant of type Rh_2 or type Rh_1Rh_2, as many as 7 anti-Rh″ sera have been observed already, two by the writer[13] and five by British investigators.[14]

These observations on competition of antigens in man may possibly find application in the prophylaxis of hemolytic disease of the fetus and newborn. While specific desensitization of the mother by injections of purified Rh haptens has not proved practicable to date, it is possible that counter-immunization of the mother during pregnancy with a potent but innocuous vaccine may serve to suppress the formation of Rh isoantibodies, and in that way prevent or diminish the severity of the disease in the fetus.

7 For nomenclature, see Wiener, A. S., *Science*, 1944, **99**, 532.

8 Wiener, A. S., *Am. J. Clin. Path.*, in press.

9 Wiener, A. S., PROC. SOC. EXP. BIOL. AND MED., 1944, **56**, 173.

10 Race, R. R., *Nature*, 1944, **153**, 771.

11 Wiener, A. S., Davidsohn, I., and Potter, E. L., *J. Exp. Med.*, 1945, **81**, 63.

12 Wiener, A. S., and Sonn, E. B., *J. Immunol.*, 1943, **47**, 461; Wiener, A. S., unpublished observations.

13 Wiener, A. S., unpublished observations.

14 Loutit, J. F., personal communication.

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Pituitary-Adrenal Cortical Control of Antibody Release from Lymphocytes. An Explanation of the Anamnestic Response.*

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The lymphocytes of normal rabbits contain a globulin identical with the normal serum gamma globulin of the rabbit.[1] Labeled globulin, *i.e.*, antibody protein, was demonstrated in lymphocytes obtained from lymphoid tissue of immunized mice.[2] The

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† Alexander Brown Coxe Memorial Fellow.

1 White, A., and Dougherty, T. F., *Abs. Proc. Meetings of Am. Chem. Soc., New York, September*, 1944; *Endocrinology*, in press.