Special Article

RECENT DEVELOPMENTS IN ISO-IMMUNIZATION BY THE RH FACTOR

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THE observation in 1939 that fetal blood may immunize the mother—in the same way, for example, that male patients may be immunized by repeated blood transfusions—paved the way for the description of the pathogenesis of erythroblastosis fetalis.[1] In a series of papers (1941 to 1942),[2-4] evidence was produced to show that (1) erythroblastosis fetalis is the result of prolonged intrauterine reaction of maternal immune agglutinins and susceptible fetal blood, (2) at least 90 per cent of all mothers of erythroblastotic infants are Rh– and their husbands and the affected infants are Rh+, (3) in the smaller group of Rh+ mothers, finer differences within the Rh complex as well as several other blood factors other than Rh (such as the blood factor of Levine and Polayes, Hr, A, B, and perhaps others) may be responsible for iso-immunization of the mother, (4) Rh– immunized mothers can be safely transfused with Rh– blood, and (5) the chances for survival of the affected Rh+ infants of Rh– mothers are better if they are transfused with Rh– blood.

Curiously enough, the presence of anti-Rh agglutinins, which is direct proof of immunization, could be demonstrated only in about 50 per cent of the Rh– mothers even if tested soon after delivery.[3] This difficulty has now been met with the demonstration independently by Race[5] in England and Wiener[6] that many Rh– mothers of erythroblastotic infants have anti-Rh antibodies which specifically unite with and coat the surface of Rh+ blood, without inducing the visible effect of agglutination. In other words, the antibody is incomplete because only the first stage of the reaction, i.e., specific union, occurs. The specifically coated Rh+ blood is now incapable of reacting with potent anti-Rh agglutinins. Accordingly, Wiener called these "blocking" antibodies while Race used the term "incomplete'" antibodies. These significant findings were soon confirmed by several workers (Diamond, Fisk and Morrow, and Levine). At any rate, it is certain that in vivo the specific reaction goes to completion so that the end result is hemolysis. In other words, clinicially, one cannot differentiate erythroblastotic infants of mothers who have anti-Rh agglutinins from those whose mothers have the incomplete antibody. By the same token, both groups of mothers are subject to severe and even fatal hemolytic reactions if they are tranfused with Rh+ blood.

Early in the work on transfusion reactions and on erythroblastosis fetalis, it was observed that not all anti-Rh agglutinins are of identical specificity.[3, 7-9] This difficulty was soon solved when it could be shown that a particular anti-Rh serum, containing but one agglutinin acting on 85 per cent of all white individuals, gave the highest value of Rh– reactions among the mothers of erythroblastotic infants.[8, 10] Levine[2] recommended a single genetic theory in order to explain the contrasting obstetric histories, i.e., either repeated or sporadic

etal and neonatal morbidity. In the former instance, the father is homozygous (RhRh) for the Rh factor, while in the latter he is heterozygous (Rhrh).

The varieties of anti-Rh sera indicated a complex or mosaic structure of the Rh factor, but no attempt was made to present the serologic details to clinicians since one serum alone served as a diagnostic reagent to detect more than 90 per cent of all individuals immunized by either pregnancy or, by the same token, repeated blood transfusions.

Wiener,[11] in an effort to elucidate the genetic theory of the Rh complex, proposed a terminology and in rapid succession several modifications thereof. At the same time, the British workers, Race[12] and Murray,[13] suggested at least two alternative terminologies so that the present situation is utterly confusing, not only to clinicians but also to immunologists and geneticists. Very briefly stated, Levine and his co-workers had shown that two common varieties of human anti-Rh sera describe four types of bloods.[3, 8, 9] Later, Race[14] and Wiener[15] described a third variety which agglutinates the blood of 30 per cent of white individuals. This serum further divides the four types, thus making eight subtypes. Fortunately, as will be shown below, it is wiser and safer for the obstetrician and clinician to consider only the clinically most important variety which gives 85 per cent reactions in a random white population. From a practical standpoint, he must know whether his patient is Rh+ or Rh– with this serum. For this reason, it becomes necessary to identify it, and the writer would like to suggest the term "diagnostic"* anti-Rh serum.

Further support for the prominent role of the "diagnostic" anti-Rh is derived from the direct correlation of Rh– individuals in several races studied and the incidence of erythroblastosis fetalis.[8, 16] Erythroblastosis fetalis is almost unknown among Chinese, because instead of 15 per cent of Rh– individuals, as in the white race, the value among Chinese is less than 1 per cent. Such correlations cannot be obtained in corresponding studies with other varieties of anti-Rh sera.

For the exceptional 8 per cent† of the mothers who are Rh+, it is necessary for the clinician to refer the blood specimen to serologic specialists in the field. Evidence for iso-immunization in these cases can be obtained by tests with (1) blood grouping sera, (2) two other varieties of anti-Rh sera reacting with 70 per cent and 30 per cent, respectively, of white individuals, (3) anti-Hr serum,[17] and rarely, still other sera. The report in these cases can be stated as an incompatibility either of a particular blood property or due to finer differences of the Rh factor. Accordingly, it seems unnecessary, at least for the present, to urge the clinician to commit to memory a series of complex terminologies, especially when his own laboratory will have only the diagnostic serum for testing.

Until all varieties of anti-Rh sera become generally available to hospital laboratories, one may expect a very occasional intra-group transfusion accident due to iso-immunization by the Hr factor or by finer differences of the Rh factor. The rule in these cases is to select donors of the same subtypes as the immunized patient. By the same token, the erythroblastotic infants of Rh+ mothers should receive transfusions of compatible blood of the same subtype as the mother. In general, mother's blood, if incompatible, may be used for her affected infant

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*This term is preferable to "standard" anti-Rh sera which refers to the experimental serum of Landsteiner and Wiener. Since this serum did not give satisfactory agglutination reactions, it was not clear whether or not it contained more than one antibody.

†The initial value of 10 per cent can probably be lowered by 2 per cent, for, in practically all cases in which the diagnosis of erythroblastosis fetalis is in doubt, the mother is Rh+. In one of Dr. Burnham's cases, it was later shown that the infant had Mediterranean (Cooley's) anemia with onset of symptoms in the neonatal period.

provided that the plasma be removed and replaced with saline or compatible plasma.

Actually, all blood specimens in selected cases (fetal and neonatal morbidity, complications of pregnancy, and intra-group transfusions reactions) should be subjected to intensive studies with all varieties of agglutinins. Since this cannot be carried out in the average hospital laboratory, the same material should be sent to qualified workers for a more intensive evaluation such as standardization of anti-Rh or other agglutinins, blocking or incomplete antibodies, and various titrations. If potent anti-Hr sera are available, tests should be made for the selection of these Rh+ fathers of erythroblastotic infants who are homozygous.[10, 18] The bloods of these individuals lack the Hr factor, and will, therefore, not be clumped by potent anti-Hr sera. The intensive study of this vast selected material should result in an increase in the supply of anti-Rh sera of all varieties. Certainly, every obstetrician is aware of the current difficulty in meeting the demands of hospital laboratories for the "diagnostic" anti-Rh serum. The situation will be much relieved as soon as potent anti-Rh sera can be produced in the experimental animal.

Obviously, it is important to determine the exact genetic behavior of the several Rh genes, but a complete theory cannot be offered unless potent specimens of the rare reagents, one variety of anti-Rh serum (30 per cent reaction in white) and the anti-Hr serum become available. Only then will it be possible to accumulate data on complete family and racial studies, the analysis of which should yield the correct genetic theory. Large scale investigations are essential since some of the subtypes are exceedingly rare.

The widespread publication of genetic schemes and terminologies for the numerous subtypes gives the clinician the wrong impression that bloods can, at all times, be classified in terms of this terminology. The fact is, however, that not even the several serologists active in this field have a constant supply of the necessary reagents for the differentiation of the several varieties of Rh+ and Rh– blood.

Undoubtedly, in time it may become necessary to adopt one of the several varieties of terminologies, but this should be decided upon preferably by an international committee as was done in the case of the four blood groups. Meanwhile, the isolated expert will perforce continue to use one of the terminologies in his own studies. However, his report to the clinician on the group of Rh+ mothers can be worded as incompatibilities detected by particular anti-Rh sera, anti-Hr sera, or blood grouping sera. Under such an arrangement, the clinician need not trouble himself with the identification of the particular variety of Rh+ and Rh– blood.

As indicated above, one may recommend to the clinician a simple genetic theory based on the behavior of the diagnostic serum, which contains but a single antibody. Accordingly, there are three genotypes, RhRh (homozygous), Rhrh (heterozygous), and rhrh (recessive). The first two represent Rh+ bloods which cannot be differentiated with anti-Rh sera. However, the Rh+ individual whose blood is not agglutinated by anti-Hr sera is homozygous for the Rh factor. A more detailed analysis which requires the use of other anti-Rh sera can be supplied by the serologic specialist.[10]

In the past three years it has become increasingly clear that once an Rh– individual is immunized, either by repeated transfusions or by pregnancies, he or she must be considered as remaining immunized for the remainder of his or her natural lifetime.[19] Thus, a woman who delivered an erythroblastotic in-

tant eleven years previously has tolerated one transfusion of Rh+ blood, but the following transfusion with Rh+ blood resulted in a violent reaction and anuria. By the same token, an Rh– woman was deprived of an opportunity of having one or two normal Rh+ children because, as a child of 6 years, she was already immunized by several blood transfusions. It is significant that in this case the first born had the most severe form of erythroblastosis fetalis, i.e., fetal hydrops. Several cases of this sort observed recently will be published in the near future.[20] Accordingly, it is necessary to consider the advisability of introducing in pediatrics the routine practice of carrying out Rh tests in all female patients to be transfused. In transfusing an Rh– female infant, it is essential to keep in mind her future pregnancies. In a broader sense, no girl or woman, regardless of age, should be transferred unless tests for Rh are carried out. Those found to be Rh– must receive only Rh– blood. With the availability of larger amounts of anti-Rh sera, such tests could be carried out in all cases, but in instances of repeated transfusions in the absence of pregnancy there is always a warning symptom of safety, i.e., a severe chill or slight jaundice which should serve as a definite indication to carry out Rh tests before another transfusion is given.

In the several reviews of the subject, no mention is made of the most significant fact that erythroblastosis fetalis establishes a precedent for fetal or neonatal morbidity attributable to a difference in a blood factor which has a normal distribution in any race. Since the blood factors involved are inherited, erythroblastosis fetalis can be considered as a disease due to genetic and constitutional causes. There already is some evidence that the antigenic factors in fetal blood other than Rh, such as A and B, may also immunize the mother with the end result of fetal and neonatal morbidity other than erythroblastosis fetalis.[21, 22]

From the very beginning of his studies, the author has stressed that the same statistical approach can be applied to any complications of pregnancy and the neonatal period.[23] Recently, Yannet[24] and Snyder[25] observed a somewhat greater incidence of Rh– reactions among 122 mothers of feebleminded infants. Further studies are required to elucidate the relationship of this group of feeblemindedness to kernicterus.

As for the mechanism of iso-immunization, it is thought that unbelievably minute amounts of fetal red blood cells in one form or another find their way into the maternal circulation in every normal pregnancy.[19] Iso-immunization occurs, however, in only a small proportion of Rh– women whose husbands are Rh+. The obstetrician is in the strategic position of being able to reassure those Rh– women who have become unduly alarmed as a result of incomplete information acquired from popular accounts in newspapers and magazines. The Rh– woman should be instructed that most of them can have two or more normal infants before they become sufficiently immunized to have an erythroblastotic infant. There is sufficient evidence to support a statement that the chances for survival of the affected infants are much better at present because the condition can be anticipated and treatment instituted promptly.

Although the silent process of iso-immunization by the fetus cannot be controlled, nevertheless the outlook for Rh– women is now much better than previously. In the first place, one can prevent the deliberate iso-immunization of the Rh– female population by transfusions with R+ blood.[26] Secondly, the blood of the pregnant Rh+ woman should be tested periodically to detect the earliest onset of iso-immunization. In general, failure to demonstrate antibodies throughout the pregnancy make it possible to give a good prognosis. If

antibodies are present and on the increase, the physician can make the diagnosis of erythroblastosis fetalis well in advance of the delivery. Premature induction of labor in selected cases in order to shorten the period of intrauterine hemolysis seems a logical procedure, but its value is still to be determined. At any rate, the affected infant should be transfused immediately with random Rh– blood via the cord vein as suggested by Mayes.[27]   No further pregnancies should be recommended until after the disappearance of all residual agglutinins or incomplete antibodies produced in the previous pregnancy. In addition to this, another interval of one or more years should elapse before another pregnancy is attempted. In this manner, it is hoped to reduce the intensity of the iso-immunization in the following pregnancy. In all cases, complete blood studies of the mother, father, and surviving children, if any, should be carried out. This will make it possible to determine the genotype of the father, whether homozygous or heterozygous.

Finally, organized effort should be directed toward the training of qualified laboratory workers in the theoretical as well as practical aspects of this new and clinically important field of medicine.

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*In twenty-three cases of erythroblastosis fetalis in the first born, a transfusion history was obtained in sixteen instances. The chances for fetal death in the first born are ten times greater in previously transfused mothers than in the control group not transfused.