THIRTY-EIGHT CASES OF HAEMOLYTIC DISEASE OF THE NEWBORN

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The following is a report of the clinical and laboratory findings and the therapeutic results in 38 cases of haemolytic disease of the newborn which were observed at the Rothschild-Hadassah-University Hospital during the years 1946-52.

Material and Methods

Thirty of the 38 cases were the result of Rh sensitization, and eight were due to incompatibility in the ABO system.

Prenatal Investigations. Pregnant women attending the health centres, which combine the functions of prenatal and infant welfare clinics, were examined routinely for Rh blood groups. The Rh-negative women were tested for Rh antibodies. Cases with Rh antibodies were invited to attend for monthly control titrations during the second trimester, and twice a week during the last trimester of pregnancy.

In a similar way serial antibody determinations for anti-A or anti-B were carried out during pregnancy in all cases with a previous history of iso-immunization and erythroblastotic infants due to ABO incompatibility. The blood group and Rh type of the father's red cells were also determined in all cases of suspected iso-immunization.

Rh antibodies in maternal sera were examined in saline and plasma. In the second part of this study the Coombs indirect method was also used. In several instances trypsinized cells were examined (Morton and Pickles, 1947).

Neonatal Investigations. Cord blood was studied in those cases in which the past history of the mother or the laboratory studies during pregnancy suggested the possibility of haemolytic disease of the newborn infant. In all cases not preceded by prenatal examinations the infant's venous blood was tested immediately following the first appearance of signs of haemolytic disease. The examinations included blood group and Rh type determinations, the direct Coombs test and a blood count.

Method of Treatment. In previous years cases of erythroblastosis foetalis have been treated by simple transfusions—single or multiple—of Rh-negative blood, or of group O-blood in instances due to ABO incompatibility.

Since 1950 exchange transfusion has been carried out in all cases in which the previous history and the clinical and laboratory findings suggested the possibility of a severe form of haemolytic disease in the newborn infant. For exchange transfusion, generally, the technique as recommended by Diamond (1947) and Mollison, Mourant and Race (1948) was used. A plastic catheter was introduced into the umbilical vein and the blood was exchanged by the 20 ml.-out 20 ml.-in method. If more than 12 hours had elapsed after birth the vena cava was entered through the saphena magna vein in the groin and the femoral vein according to the technique described by Arnold and Alford (1948).

In two instances, owing to the technical difficulties, the umbilical route had to be abandoned for the withdrawal of blood. The remainder of the blood was withdrawn from the sagittal sinus through the anterior fontanelle. In general, from 400 to 500 ml. of donor blood was used for replacement, leaving a surplus of about 50 ml. of transfused blood above the amount of withdrawn blood.

I. Rh Incompatibilities

Incidence. There were 29 cases of haemolytic disease of the newborn among 10,800 deliveries, an incidence of 1 in 372, during the years 1946-52, excluding 1948. For the year 1948 records were incomplete as a result of war conditions. In this year one case of haemolytic disease was found and is not included in the incidence calculation but is included in this series.

This relatively low incidence rate may have been due to the fact that primiparas were particularly selected for delivery at our maternity ward and

represented, therefore, a disproportionate percentage of the total number of deliveries. Previously Gurevitch, Brzezinski and Polishuk (1947) reported an incidence of 1 in 2,100 deliveries among Jerusalem Jews. The present apparently increased incidence of 1 in 372 may be attributed to expanded facilities for laboratory diagnosis.

Sex. There were 16 male and 14 female infants. Eleven (69%) of the male infants and 11 (79%) of the female infants recovered. Although this number of cases is too small to permit conclusions, no significant difference of mortality and recovery rates in both sexes was observed.

Past History of the Mother. Twenty-nine of 30 erythroblastotic infants were born to multiparas, and only one case—a stillborn infant—was the result of the first pregnancy of an Rh-negative woman. In this case no transfusion or blood injection was known to have been given to the woman previously. A titre of 1 in 512 for saline anti-Rh antibodies was found in the mother's serum shortly after delivery. Two years later the same woman gave birth to an infant severely affected by haemolytic disease.

In one case a blood transfusion appeared to have acted as the first sensitization against the Rh factor, and was followed by stillbirth after a first pregnancy and an erythroblastotic infant after the second pregnancy.

One case appeared to be an instance of late sensitization, since the erythroblastotic child, born of the fourth pregnancy, was preceded by three normal unaffected siblings. In this instance complete recovery occurred without any treatment. Allen, Diamond and Vaughan (1950) have suggested that such cases of 'late sensitization' show a milder course of the haemolytic disease.

Jaundice. Except for three mild cases, almost all liveborn infants in this series exhibited jaundice during the first 24 hours after birth. The exact time of appearance of jaundice was as follows:

At birth	..	6 cases
4-12 hours after birth	..	9 "
12-24 hours after birth	..	6 "
Second day	..	2 "
No jaundice	..	3 "

It is of interest that six infants had jaundice at birth. In the series reported by Diamond, Vaughan and Allen (1950) there was no case of jaundice at birth.

The three infants without jaundice were mild cases with signs of anaemia only, so-called 'congenital anaemia'. Two of them recovered spontaneously, the third after a simple transfusion.

Spleen and Liver Enlargement. The spleen was found enlarged (over 2 cm. below the costal margin) in 18 cases, normal in three. The liver was enlarged

in 12 cases, normal in nine. In five cases exact data on the size of spleen and liver were not available. In the remaining four cases the infants were stillborn.

Direct Coombs Test. Of the 19 cases examined by the antiglobulin test, 14 showed positive results. The first three cases yielded negative results which may have been due to inadequate technique at the beginning in using the Coombs test. Two other cases, siblings, presented the problem of double incompatibility.

Anaemia. Blood counts throughout the whole series ranged from 1,300,000 to 4,500,000 red cells per c.mm., and from 6.5 g. to 15 g. of haemoglobin per 100 ml. of blood, with the majority of cases on the lower side of the range. An abnormally elevated number of nucleated red cells was found in all but three cases.

No definite correlation between the degree of anaemia or the number of nucleated red cells and the severity and prognosis was found.

Serological Findings in Maternal Sera. An analysis of the serological findings revealed no clear correlation between the titre of Rh antibodies in the maternal sera and the severity of haemolytic disease in the newborn infant. Thus, two cases suffered from mild haemolytic disease in presence of a high antibody titre, while another case resulted in the death of the affected infant in the presence of a relatively low antibody titre in the mother's serum. It is noteworthy, however, that all cases of stillbirth in this series were accompanied by especially high antibody titres in the respective mother's serum.

A special problem is represented by the cases of double incompatibility in the Rh- and ABO-system at the same time. These cases were characterized by the absence of Rh antibodies and by low anti-A or anti-B titres in the maternal serum. Despite these findings, the typical picture of severe haemolytic disease existed in the infants, and in one case resulted in kernikterus and death of the infant. The inhibition of antibody formation may possibly be due to 'competition of antigens' as suggested by Wiener (1945). It is of interest that the Coombs test was also negative in two of these cases.

II. ABO Incompatibilities

A review of the literature on this subject showed that up to now no clear-cut opinion on the validity of 'ABO erythroblastosis' has been reached. Thus Pickles (1949) believed that 'only very stringent tests for incomplete antibodies anti-A and anti-B, and the exclusion of other haemagglutinins in each case can give unequivocal proof of ABO erythroblastosis'. Zuelzer and Mudgett (1950) were of the opinion that a rise in anti-A or anti-B titre and the detection of

FINDINGS IN EIGHT CASES OF ERYTHROBLASTOSIS

Year	Case	Sex	Pregnancy	Previous History and Sensitization of Mother	Clinical Findings			Blood Group M = mother I = infant F = father
					Jaundice	Enlarged Spleen	Enlarged Liver	
1947	1	F.	4	2 normal children 1 stillbirth	2nd day	—	—	M: OCDe/cde I: ACDe/cde F: ACDe/cde
1950	2	F.	3	2 normal children	Stillbirth			M: OCDe/cde I: ? F: ACDe/cde
	3	M.	3	1 abortion 1 premature infant, died	2nd day	+	—	M: OCDe/CDe I: ACDe/CDe F: ACDe/CDe
1951	4	M.	4	See case 3 above; induction of labour 39th week	At birth	—	—	M: OCDe/cde I: ACDe/cde F: ACDe/CDe
	5	F.	6	1 child died immediately after birth, 2 children died at later age 2 normal children	5 hours	+	+	M: OCDe/cDE I: ACDe/cde F: BCDe/cde
	6	F.	1		8 hours	—	—	M: OCDe/cde I: ACDe/cde F: ?
	7	M.	1		10 hours	—	—	M: OCDe/cde I: ACDe/CDe F: ?
1952	8	M.	1		At birth	+	—	M: OCDe/cde I: BCDe/cde F: BCDe/cde

incomplete antibodies were not sufficient criteria to establish the validity of ABO erythroblastosis, as they could be found commonly in normal hetero-specific pregnancies. On the other hand, Wiener, Wexler and Hurst (1949) were impressed by the growing number of well documented reports of typical and sometimes fatal cases as evidence for AB sensitization in the aetiology of haemolytic disease of the newborn.

Despite the difference of opinion on this subject, and although we feel that doubts are justified as long as no more accurate proof of the validity of ABO erythroblastosis can be established, our series adds another eight cases of typical haemolytic disease of the newborn in which no Rh incompatibility existed and in which AB sensitization appeared to be the probable aetiology. Other factors, however, could not be completely excluded, as we were not equipped with facilities for investigations of other antigenic systems.

Findings in the ABO group of cases are summarized in Table 1.

With regard to the past history of mothers, the composition of this group was different from the Rh group. Four of the eight cases of haemolytic disease occurred as a result of first pregnancies and

without a history of previous blood transfusion. These cases, therefore, exemplify the peculiarity of 'ABO erythroblastosis' which occurs occasionally in first pregnancies (Pickles, 1949).

Jaundice appeared immediately at birth in two of the cases, and up to the second day in the remainder of cases. The Coombs test was negative in all of our cases. The inadequacy of the Coombs test for the detection of univalent anti-A or anti-B has been repeatedly stressed by various authors, and remains as yet unexplained (Coombs, Mourant and Race, 1946; Wiener *et al.*, 1949). Progressive anaemia and the presence of many nucleated red cells in the peripheral blood was a common feature of our cases.

As seen from Table 1, incomplete anti-A or anti-B titres in maternal sera ranged from 2,000 to 128,000 units in plasma. While Smith (1945), Tovey (1945) and Zuelzer and Mudgett (1950) have pointed to the fact that high titres may occur in normal hetero-specific pregnancies without any pathological effect on the foetus, Wiener *et al.* (1949) have shown a much higher range of titre in cases of affected infants as compared with a series of unaffected infants from heterospecific pregnancies.

Although it seems established that high univalent anti-A or anti-B titres *per se* are not necessarily

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FOETALIS DUE TO ABO INCOMPATIBILITY

Laboratory Findings					Treatment	Results	Remarks	
Infant				Maternal Anti-Bodies (in units)				
Direct Coombs	R.B.C. (in millions)	Hb. (g.)	Nucleated R.B.C. (per cent. of total nucleated cells)	Saline				Plasma
?	2.8	8	50	anti-A 16,000	32,000	Simple transfusion at 4 weeks	Died	Late admission to hospital
?				anti-A 2,000	16,000		Stillbirth	
—	2.6	15	0	anti-A 1,000	4,000	Simple transfusion on fourth day	Died (kernikterus)	Late admission to hospital
—	5.5	15	30	anti-A 2,000 anti-C 0	16,000 0	Exchange transfusion	Recovered	Induction of labour 39th week
—	4.2	12	37	anti-B 128	2,000	Simple transfusion	Recovered	Premature birth (2,300 g.)
—	2.6	9	40	anti-A 256	128,000	Simple transfusion	Recovered	
—	3.8	12.5	0	anti-A 512 trypsin cells 2,000	512	Simple transfusion	Recovered	
—	2.7	11.5	11	anti-B 512 anti-E 0	1,000 0	Simple transfusion	Recovered	

correlated with a diseased infant, only such high titres may warrant the designation of cases as haemolytic disease possibly due to ABO incompatibility. In view of this criterion, cases with lower titres were not included in this series. Of the fatal cases in this group, one was a stillbirth, and two died after inadequate treatment (Nos. 1, 3). The mother of cases Nos. 3 and 4 was of special interest. During her pregnancy in 1950 (Case 3) her plasma titre of anti-A antibodies was 4,000. The infant of this pregnancy died of haemolytic disease. In the following year, during another pregnancy (Case 4) the plasma titre of anti-A antibodies rose to 16,000. Labour was induced in the thirty-ninth week of gestation. Despite the increased titre of anti-A antibodies and the increased severity of the haemolytic disease in Case 4, the infant recovered following exchange transfusion.

III. Treatment

The indication for treatment was based on clinical and haematological grounds and was independent of whether the underlying cause was found to be Rh or ABO incompatibility.

Of the 33 liveborn infants, 16 were treated by simple transfusions, 13 by exchange transfusion and

four mild cases recovered spontaneously. Of the group of 16 infants who received simple blood transfusions, five died. The 13 infants treated by exchange transfusion all recovered despite the fact that the more severe cases were selected for this treatment (Table 2).

TABLE 2
RESULTS OF TREATMENT IN 33 CASES OF HAEMOLYTIC DISEASE OF THE NEWBORN

	Simple Transfusion	Exchange Transfusion	No Treatment
Recovered ..	11	13	4
Died ..	5	—	—
Total ..	16	13	4

The difficulty of evaluating and comparing various methods of treatment of haemolytic disease of the newborn has been repeatedly pointed out (Vaughan, Allen and Diamond, 1950). It is primarily due to the fact that the early estimation of the severity of a case of haemolytic disease is usually most difficult. Nevertheless from our figures, though small, the conclusion may be drawn that exchange transfusion proved to have a definite advantage in the treatment of haemolytic disease of the newborn, and represented therefore the treatment of choice for all severe

cases of this disease. It has to be borne in mind, however, that in three cases out of the five deaths in the group of simple transfusions treatment was started relatively late, and that early simple transfusion perhaps might have changed the final outcome of these cases.

The value of exchange transfusion for severe cases and the adequacy of early, simple transfusion for milder cases has been convincingly worked out in the recent controlled trials of treatment by Mollison and Walker (1952).

Induction of labour in our series was performed in two cases only, in both of them because of the history of previous fatal haemolytic disease in several siblings, and rise in antibody titre in the mother's serum during pregnancy. In both instances labour was induced in the thirty-ninth week of gestation, and resulted in a living erythroblastotic infant who recovered after exchange transfusion.

Summary

Clinical, haematological and serological findings, and the therapeutic results in a series of 38 cases of haemolytic disease of the newborn are reported. Of

these, 30 were due to Rh incompatibility, and eight to ABO incompatibility.

The problem of ABO incompatibility in the aetiology of haemolytic disease of the newborn is discussed.

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