ATTEMPT TO PREVENT ERYTHROBLASTOSIS FOETALIS BY USE OF CORTISONE DURING PREGNANCY

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Since the discovery by Landsteiner and Wiener (1940) of the rhesus factor great advances have been made in the understanding of erythroblastosis foetalis, and it is now firmly established that the condition results from the development in the pregnant woman of immune antibodies capable of crossing the placental barrier and causing excessive red-cell destruction in the foetus. This recent knowledge has led to several attempts to inhibit the development of such antibodies. Thus Wiener and Sonn (1946) gave injections of typhoid and pertussis vaccines during pregnancy in the hope that the administration of a strong antigen might interfere with the development of antibodies in response to the weaker (Rh) antigen. Kariher (1947), acting upon reports that ethylene disulphonate was alleged to be of value in allergic conditions, administered this substance in minute amounts to pregnant women who had previously given birth to infants with haemolytic disease. His results suggested that such treatment may have some effect in lowering the antibody titre in the maternal serum. Carter (1947, 1949) claimed to have isolated from human Rh-positive red cells a fraction containing an Rh hapten, and reported that injections of this fraction into Rh-sensitized pregnant women resulted in a fall in titre of the circulating Rh antibodies. Other workers, notably Howe and Rustigian (1950), and Spurling *et al.* (1950), have failed to produce results comparable to those of Carter. So far no widely accepted therapeutic procedure has resulted from these attempts to inhibit Rh antibodies.

At the present time the effects of cortisone and A.C.T.H. upon immunity reactions are incompletely understood. The available reports indicate a species difference: cf. guinea-pig (Dews and Code, 1951), rabbit (Germuth and Ottinger, 1950), and man (Mirick, 1951). In man A.C.T.H. and cortisone apparently do not inhibit the development of antibodies (Havens *et al.*, 1951) but are able to prevent hypersensitivity reactions (Long and Favour, 1950). It seems that in man the adrenal agents prevent either the union of antigen and antibody or the tissue injury which results from such union.

Among the numerous trials of cortisone and A.C.T.H. in human disease, those which seem most relevant to the present investigation concern the condition of "idiopathic" acquired haemolytic anaemia, in which the haemolytic process is associated with a positive direct Coombs reaction. Here, it seems reasonable to assume that the patient develops auto-antibody which, attaching itself to the red cells, is responsible for the increased rate of their destruction. Dameshek (1950) reported that

A.C.T.H. had a beneficial effect in two such cases which had failed to respond to splenectomy. Gardner (1950) claimed improvement in three further cases treated with A.C.T.H.; he thought that there was temporary suppression of antibodies active against the patients' red cells, the strength of the direct Coombs reaction being diminished. Davidson *et al.* (1951) also reported a good response to A.C.T.H. in such a case. More recently, Clearkin (1952) reported the effect of A.C.T.H. on two cases of chronic acquired haemolytic anaemia. In one case there was no noticeable effect; in the other the haemolytic process was greatly diminished, although the red cells reacted more strongly than before with Coombs reagent. This last observation suggests that A.C.T.H. may have influenced the haemolytic process not by inhibiting antibody formation but by diminishing the rate of destruction of red cells coated with auto-antibody. There is thus evidence that A.C.T.H. may have a beneficial effect in cases of haemolytic anaemia associated with the presence of auto-antibody attached to the red cells. The purpose of the present study was to determine if cortisone given during pregnancy would prevent or diminish the severity of haemolytic disease in the foetus.

It was thought that a case suitable for such a trial should present the following features: (1) a history of erythroblastosis foetalis in a previous infant or infants; (2) a recent pregnancy resulting in a severely affected infant; (3) the presence of incomplete Rh antibody in high titre at an early stage of pregnancy; and (4) a homozygous Rh-positive husband. The following report concerns a case which fulfils these criteria.

Case Report

A married woman aged 33 had had two previous pregnancies. The first of these, during which she was discovered to be Rh-negative and her husband Rh-positive, resulted in the birth of a full-term normal Rh-positive male infant on March 19, 1948. In the second pregnancy, in 1949, her serum was first examined for irregular antibodies at the 32nd week, and was found to contain incomplete anti-D in a titre of 1 in 4. For this reason surgical induction was carried out at the end of the 37th week; the titre of incomplete anti-D was then 1 in 32. The infant, a premature male weighing 6 lb. 8½ oz. (3 kg.), became jaundiced within 20 minutes of birth. Examination of the cord blood showed: Hb, 48%; red cells, 2,500,000 per c.mm., Rh-positive; direct Coombs test positive. A simple transfusion of 120 ml. of Rh-negative blood was given. Jaundice deepened rapidly, and the child died 32 hours after birth. Post-mortem examination showed generalized oedema, ascites, and jaundice, and the brain was icteric. The appearances were regarded as typical of erythroblastosis, and no other abnormality was found.

On April 24, 1951, the patient reported at the antenatal clinic of Stobhill General Hospital. She was then six weeks pregnant. Her serum contained incomplete anti-D in a titre of 1 in 64. Blood grouping was carried out upon the patient, her husband, and the first child; the results are

Rh Genotype Determinations upon the Patient, Her Family, and the Husband's Relatives

	ABO Group	C	D	E	c	e	Probable Rh Genotype
Patient	A	—	—	—	+	+	cde/cde
Husband	A	+	+	+	+	+	CDe/cDE
First child	A	+	+	+	+	+	cDE/cde
Husband's father ..	A	+	+	+	+	+	CDe/cDE
" mother	O	+	+	+	+	+	cDE/cde
" brother—1	O	+	+	—	+	+	CDe/cde
" "—2	O	+	+	—	+	+	CDe/cde
" "—3	A	+	+	+	+	+	CDe/cDE

shown in the Table. In an attempt to determine the husband's Rh genotype, grouping tests were performed upon his parents and siblings. These also are shown in the Table. From the blood-group determinations it seemed highly probable that the husband was homozygous for the antigen D, and it was confidently predicted that in the present pregnancy the foetus would be Rh-positive, and would develop erythroblastosis foetalis.

Throughout pregnancy incomplete anti-D antibody was present in the patient's serum, samples of which were obtained at frequent intervals and tested with a 3% suspension of Group A, CDe/cDE red cells in 25% bovine albumin. The antibody titres are shown in Fig. 1. All tests

FIG. 1.—The titres of incomplete anti-D in the maternal serum during pregnancy and the puerperium. The titres represent the dilution of serum after addition of an equal volume of a suspension of the test red cells in 20% albumin.

were performed with red cells from the same individual; at no time was any irregular saline agglutinin detected. From the 26th week of pregnancy renal glycosuria was observed; otherwise pregnancy appeared to progress normally.

At the end of the 31st week the patient was admitted to hospital for cortisone therapy. After two weeks, during which control observations were made (see below), the patient, now at the end of the 33rd week of pregnancy, was given 50 mg. of cortisone twice daily in milk. This dose was continued until the end of the 37th week. Since cortisone given in such doses can interfere with the body's defence against infection (Kass *et al.*, 1950a; Fred *et al.*, 1951) and disturb the electrolyte pattern of the body fluids (Sprague *et al.*, 1950), careful observation of the patient was made during the course of therapy. The lungs were x-rayed before and at the end of treatment, and the urine and skin were often examined for evidence of infection. To detect undue sodium retention—which is undesirable in a pregnant woman—serial observations were made of body weight, packed cell volume, and blood pressure. An electrocardiogram was taken weekly to give warning of any significant fall in tissue potassium, a change which may result from cortisone therapy. A special diet low in sodium and rich in potassium was not given. We were also interested to observe whether the patient's renal glycosuria was aggravated by cortisone, since several authors (Conn *et al.*, 1948; Kass *et al.*, 1950b; Dustan *et al.*, 1951) have reported the occurrence of renal glycosuria during A.C.T.H. therapy.

In Fig. 2 are presented the results of these control observations. The eosinophils—47 to 75 per c.mm. before treatment—fell to below 20 per c.mm., and with minor rises remained low during cortisone administration. After parturition the dose of cortisone was gradually reduced to allow the patient's pituitary gland to recover control of her adrenal function. During this phase of diminishing cortisone dosage the eosinophils rose steadily to reach 200 per c.mm. The

weight and P.C.V. curves showed a slight divergence from each other early in the course of treatment, indicating a little salt retention, but towards the end of therapy became parallel. The blood pressure remained steady at about 110/70 mm. Hg. Weekly E.C.G.s showed no evidence of potassium depletion. Glycosuria increased slightly during the first few days of treatment.

At the conclusion of the course of treatment, the patient then being at the end of the 37th week of pregnancy, labour was induced by artificial rupture of the membranes and withdrawal of 1 litre of liquor amnii, which was not noticeably bile-stained. After a six-and-a-half-hour labour she was delivered of a live female infant weighing 6 lb. 15½ oz. (3.2 kg.). The umbilical cord was bile-stained and oedematous; the placenta weighed 500 g. and appeared normal both on naked-eye and on microscopical examination. A sample of cord blood showed: Hb, 90% Sahli (13 g. per 100 ml.); red cells, 4,100,000 per c.mm.; reticulocytes, 12%; normoblasts, 40,000 per c.mm. The serum was noticeably jaundiced (icteric index=20 units). The blood was Rh-positive and the direct Coombs test was positive. Incomplete anti-D was present in the serum in a titre of 1 in 8. The child was clinically normal at birth, but at the end of three hours jaundice became apparent. A replacement transfusion was performed by Dr. D. H. Wallace, 435 ml. of Group O Rh-negative blood being administered by the umbilical vein, and 420 ml. concurrently withdrawn. No adverse effects were noticed during the transfusion, and jaundice faded within the next week. Since then the child has remained well without further treatment.

A sample of the child's red cells was obtained at the age of 14 weeks, and was shown to be Group O, cDEe, the probable Rh genotype being cde/cDE. Some of the samples of maternal serum obtained during pregnancy and stored frozen were retested for irregular antibodies, using albumin suspensions of the child's red cells and of the red cells of the individual originally used to test these samples. The titres of incomplete antibody were the same with both cell suspensions, a result which excludes the possibility that the infant's red cells contained a D^u variant of the D antigen which might have reacted weakly with the maternal antibody.

The puerperium was uneventful. Healing of a second-degree tear of the perineum sustained during labour was not delayed.

Discussion

When seen at an early stage of her third pregnancy, our patient was known to be Rh-negative, and her serum contained Rh incomplete antibody in high titre. Her husband was most probably homozygous Rh-positive, and the previous infant, born two and a half years ago, had died of erythroblastosis foetalis in spite of simple transfusion; from these considerations it seemed probable that the present pregnancy would result in intrauterine death or in an infant

FIG. 2.—Control of cortisone in the Rh-sensitized patient during her third pregnancy.

severely affected by erythroblastosis foetalis. The maternal Rh incomplete antibody remained at a high level throughout pregnancy, which was terminated at the end of the 37th week, immediately after completion of a four-weeks course of cortisone therapy. The infant was Rh-positive, and was mildly affected by erythroblastosis foetalis. An exchange transfusion with Rh-negative blood was performed to minimize the risk of icterus gravis, and the child has since remained well. The survival of this child and the death of the previous one may be partly attributable to the different methods of transfusion therapy adopted in the two cases. However, this does not alter the fact that the previous infant was severely affected by erythroblastosis foetalis and was in poor condition at birth, whereas the present infant was less severely affected and was born in good condition.

Although it occasionally happens that erythroblastosis foetalis is less severe in successive affected infants, this is the exception and is usually associated with an interval of many years between pregnancies (Pickles, 1949). We thus consider that the severity of erythroblastosis foetalis in the case described was less than had been anticipated, and, while we fully realize that one case proves nothing, the problem of obtaining a living and healthy child from a highly sensitized mother is so difficult of solution that any measure which might contribute in however small a degree to such a result is worthy of investigation. It is in the light of such considerations that we venture to publish this case and to suggest that the result is encouraging enough to warrant further trials in Rh-sensitized pregnant women. If these trials prove the value of cortisone in protecting the foetus from the effects of maternal Rh-immunization, then the mode of action of the hormone might profitably be discussed. At present we intend to confine our remarks to the time of administration, dosage, and control of cortisone therapy.

It is well established that the damage to the foetus resulting from maternal Rh-immunization occurs during the later months of pregnancy. Induction of labour before term, however, does not increase the chances of survival of the affected infant owing to the increased risk of icterus gravis in premature infants (Vaughan *et al.*, 1950). Administration of cortisone to the mother must therefore be regarded as an attempt to protect the foetus during late pregnancy. The time to start cortisone therapy must be decided from the previous history. In our case the previous infant, born at the end of the 37th week, was severely affected by erythroblastosis foetalis, and we should have preferred to begin treatment at about the 30th week of the present pregnancy, and continue until term. However, we had only enough cortisone for four weeks' treatment, and, as we thought that treatment should continue until the birth of the infant, we delayed giving the hormone until the end of the 33rd week and induced labour at the end of the 37th week.

With regard to the dosage of cortisone, our patient received 100 mg. daily by mouth. No serious complications occurred, and the circulating eosinophils remained below 20 per c.mm. during therapy. The eosinophil level is the most sensitive indicator of glucocorticoid activity, and it may be that the dosage effecting considerable reduction of maternal eosinophils is inadequate to protect the foetus from maternal Rh antibody. However, this protective effect can be assessed only after delivery. In our case the haemolytic process was not abolished, although it may well have been diminished. We believe that in future trials larger doses, of the order of 150 mg. daily, should be given, provided that the mother is protected from the risk of serious side-effects. This necessitates careful assessment of control observations, especially of changes in blood pressure, P.C.V., and body weight.

Doerner *et al.* (1951) treated two Rh-sensitized pregnant women with cortisone in larger initial doses than we used; in both of their patients a urinary infection and a mild Cushing's syndrome developed after about two weeks of therapy, and the dosage of cortisone was thereafter reduced to 25 mg. daily. In one case hydrops foetalis developed;

in the other, the infant was anaemic at birth but survived after a simple transfusion. In this second case the infant received cortisone after birth, and in a third case cortisone and A.C.T.H. were administered solely to the newborn infant. Since erythroblastosis foetalis begins *in utero* and continues after birth, it seems logical to administer cortisone both to the mother during pregnancy and to the newborn infant. However, exchange transfusion is of well-established value in the newborn erythroblastic infant, and in our case we preferred to treat the mildly affected infant by this method rather than by further trial of cortisone.

Summary

An Rh-negative pregnant woman whose previous infant, 2½ years ago, had died of erythroblastosis foetalis was treated with 100 mg. of cortisone daily by mouth from the 34th to 37th weeks of pregnancy, when labour was induced.

No harmful effect of cortisone therapy was observed in the mother. High-titre incomplete anti-D was present in the maternal serum throughout pregnancy. The infant was mildly affected by erythroblastosis foetalis, and has remained well following an exchange transfusion.

This result is encouraging enough to warrant further trials of cortisone therapy. The criteria useful in choosing suitable cases, and the dosage and duration of such therapy, are discussed. The precautions taken to safeguard the mother from possible ill effects of cortisone are presented in detail.

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There is little chance that reception centres for tramps at present linked with hospitals or homes for old people will soon be moved to new premises. Such associations date from the days of the Poor Law institutions, and are everywhere condemned. The capital cost of segregation makes it impracticable. This unwelcome news is given in a recent report of the National Assistance Board. The report adds that there are fewer tramps than before the war. It is rare now for the nightly total of casuals at reception centres to exceed 2,500.