with a particularly alert antibody-forming system? If so, was this a contributing cause of their two obscure diseases? Or is the sensitivity secondary to the disease? Or is there no connexion between the diseases and the presence of the antibodies other than the fact that the diseases made repeated transfusions necessary, and so gave the patients a chance to demonstrate powers that may not be very uncommon?

The patient possesses antibodies to all the antigens which she lacks in five blood-group systems. The evidence for the presence of each antibody was examined statistically. Sufficient samples of red cells were tested to reduce to a very low degree of probability the chance that any of the reactions were merely coincidental.

Several of the antibodies in this serum were weak and might have disappeared on heterospecific absorption. The need for absorption could be overcome by the use of highly selected red cell samples; these were chosen from a large panel of some 400 persons grouped for the A_1A_2BO , MNS, P, Rh, Lutheran, Kell, and Lewis systems.

Since the patient lacks the antigen P it is surprising that no anti-P could be found in her serum, for this antibody is of much more frequent occurrence (Henningsen, 1949) than most of those which were identified; moreover, she must surely have received some P+ blood. In view of the demonstration by Mainwaring and Pickles (1948) that Lutheran is quite an effective antigen, it would seem possible that the patient did not receive any Lu(a+) blood.

It was rather disappointing that no "new" blood-group antibodies could be demonstrated in this serum. The search, however, was greatly limited by the rarity of blood samples compatible with the antibodies known to be present in the serum.

Summary

A patient suffering from osteosclerosis with extramedullary haematopoiesis and who had been transfused 20 times was found to have nine blood-group antibodies in her serum; these were anti-A, anti-B, anti-N, anti-S, anti-E, anti-C^w, anti-K, anti-Le^a, and anti-Le^b. Five of these antibodies are very rare.

We wish to thank Dr. L. K. Diamond, of the Blood Grouping Laboratory, Boston, for making it possible for one of us (R.S.) to work there for a time. We also thank Dr. B. H. Kirman and Mr. J. E. Stevens, of the Fountain Hospital, Tooting, for their help in setting up the large panel of control cells.

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The British Rheumatic Association, which is a voluntary body working with the goodwill of the Ministry of Health to improve the medical, social, and economic position of sufferers from rheumatism, has formed a financially independent association called B.R.A. Homes, Ltd.—a registered housing society with charitable status. The society is in need of funds to equip a Georgian crescent (St. Ann's) at Buxton, Derbyshire, with 60 amenity beds for those who cannot get urgently needed treatment in hospital but who can receive it at St. Ann's as out-patients under the Health Act. There will also be 30 furnished flatlets for elderly disabled sufferers from rheumatism needing nursing care. St. Ann's will receive patients from all parts of the country, but it is hoped to open similar homes later elsewhere. A total sum of about £50,000 is needed, and contributions will be gratefully accepted by B.R.A. Homes, Ltd., at 5, Tite Street, London, S.W.3.

MULTIPLE ANTIBODY RESPONSE TO REPEATED TRANSFUSIONS

BY

R. H. MALONE, M.D., D.P.H.

AND

J. COWEN

(From the Sheffield Blood Transfusion Centre)

As our knowledge of red cell antigens and antibodies increases so the selection of blood suitable for transfusion from one individual to another becomes an undertaking of increasing complexity. The routine cross-matching tests carried out in hospital practice are usually sufficient to detect the presence of the antigens A, B, O, and D and the corresponding antibodies when performed by trained and competent medical officers or technicians. Where, however, repeated transfusions are required the risk of immunization or a transfusion reaction due to the rarer antibodies, which are not readily detectable by such techniques, is one that cannot be ignored. In such cases cross-matching calls for special techniques, and the search for and identification of the rarer antibodies may become an arduous and complex task best suited to laboratories specially equipped for carrying out investigations of this nature.

While certain individuals are more readily immunized by blood antigens than others, it is reasonable to suppose that the greater the number of transfusions given to a patient the greater is the likelihood of his developing antibodies of one sort or another; and with the continuing popularity of multiple transfusions as a therapeutic measure in certain diseases and the introduction of new techniques for antibody detection it is becoming increasingly clear that the so-called "rare" antibodies are of much more frequent occurrence than is generally believed.

The case reported here is one of several encountered during the past few months in which "rare" antibodies have developed as a result of repeated transfusions. It illustrates multiple antibody response to the transfusion of blood from 24 donors, all of whom were "compatible" in respect of the A, B, O, and D blood antigens.

Case Report

The patient, a widow aged 64 with four normal children, had no history of miscarriages or of any transfusions of blood. She complained of lassitude, dyspnoea, and oedema of the ankles of six to eight months' duration and of passing very dark urine.

On examination on September 2, 1949, there was profound anaemia but no icterus. The red cell count was 1,280,000 per c.mm., haemoglobin 25%, serum bilirubin 3.3 mg. per 100 ml. The spleen was palpable and the liver "(?) slightly enlarged." The heart was slightly enlarged to the left, with an apical systolic murmur. A provisional diagnosis of acquired haemolytic anaemia was made.

On September 24 a specimen of her blood was received at the Sheffield Blood Transfusion Centre with a request for two pints (1.14 litres) of packed cells. Her cells were found to be Group A Rh-positive, and no difficulty was encountered in matching her serum against the cells of four D-positive donors in saline and albumin suspension. Two pints of packed cells were transfused: the haemoglobin rose to 45%; the temperature, which had been swinging wildly up to this stage, settled down and she seemed very much better. She was transfused again on September 30 with good results.

Fourteen days later a further specimen was received and her cells were found to be A₁ D NNp. Her serum was tested against a panel of 10 Group O cells of various Rh types in saline and albumin at 37° C., room temperature, and 4° C., and an indirect Coombs test was performed. These tests were all negative, indicating that no Rh or cold agglutinins had been formed. The patient's serum was tested against her own cells at 4° C., room temperature, and 37° C., and a direct Coombs test was performed upon her washed cells. All of these were negative and no haemolysins were detected.

On October 16 her temperature again became irregular and definite icterus appeared. The spleen was much larger than before and the haemoglobin had dropped to 32%. Next day a further specimen of blood was received with a request for more packed cells, the hospital having reported difficulty in cross-matching. Four bottles were matched, two pints of whole blood dispatched as an emergency on the night of the 17th, and a pint (570 ml.) of packed cells next morning. The transfusion failed to stop the haemolytic process.

Further investigations were then undertaken on the sample of blood received on October 17. She was found to be Group A₁, NNS, ?P, R₁R₁ (CDe/CDe); Lewis, Lutheran, Kell, and Willis negative. It was suspected that the doubtful reaction with anti-P serum was due to transfused cells carrying the P antigen, as she had previously been found to be P-negative. The serum, which was very yellow (a condition not previously observed), was tested further for antibodies. On this occasion there was a visual positive reaction with Group O R"R" (cdE/cdE) cells when incubated at 37° C., and in addition a positive reaction at 4° C. and room temperature against her own cells and against each of 16 O, two A₁, and two A₂ cells of various Rh types and including M positives and negatives and P positives and negatives. Weak reactions occurred with two cells lacking the M and E antigens. Doubtful reactions were noted when the patient's serum was placed against her own washed cells, and the direct Coombs test was weakly positive. It was difficult to decide whether this reaction was due to autoagglutinin or sensitized donor cells in the patient's circulation or to the haemolytic anaemia itself, but, in view of the weakness of the Coombs reaction and the doubtful autoreaction, the presence of donor cells was thought to be the more probable explanation. The serum definitely contained anti-E together with one or more antibodies reacting strongly with saline suspensions of all cells in the panel at 4° C. and room temperature, and weakly at 37° C. with some cells possessing the M but not the E antigen. Some strongly positive reactions occurred in albumin at 37° C., but only in the presence of the E antigen, and it was possible to distinguish by the strength of the reaction cells with a single dose of E from those with a double dose. The indirect Coombs test was positive with R₂R₂ (cDE/cDE) and R"R" (cdE/cdE), but negative with R₂r (cDE/cde) cells.

E-negative packed cells were transfused on October 22, 28, and 30. Cross-matching was performed as before, using the saline, albumin, and Coombs methods, but it had become increasingly difficult to find cells which would match with her serum by all three methods.

These transfusions were without beneficial effect apart from increasing the haemoglobin content of the blood, and by October 28 the spleen had greatly enlarged and there was a marked increase of urobilinogen in the urine. Her general condition had deteriorated considerably and her heart was failing rapidly. She died on October 31. There was no evidence of a transfusion reaction after any of the transfusions given during her stay in hospital.

A post-mortem specimen of her blood was tested with anti-M, anti-N, and anti-P sera, and a positive reaction was obtained with each. With anti-M and anti-P, however, the clumped cells lay in a negative field, and since the patient's cells were originally typed as NNp it is almost certain that the clumping was due to donor cells. The specimen was allowed to stand at 4° C. for some hours in order to absorb any autoagglutinin that might be present, and the absorbed serum was tested against a panel of 12 Group O cells of various Rh and MNP types. The anti-E

was again in evidence at 37° C. in both saline and albumin cell-suspensions and had a titre of 1 against cells with a single dose of E (NNR₂r) and 8 against those with a double dose (NNR₂R₂). It was also noted that at 37° C. the serum agglutinated saline suspensions of cells carrying the M antigen, irrespective of the presence or absence of E: in albumin, however, only the cells bearing E were agglutinated. At room temperature only two types of cells in the panel were not agglutinated—NNP and NNp—both E-negative. From these findings it was evident that anti-M had been formed. The titre of the anti-M at room temperature was found to be 4. At 4° C. the cell type NNp was agglutinated while NNp was not affected. This indicated that the serum contained anti-P in addition to anti-M and anti-E. The patient's serum was finally placed against her own cells, and while no reaction took place at 37° C. agglutination did occur at 4° C., the clumps appearing in the negative field; this is not surprising, since the specimen was to some extent composed of donor cells and gave positive reactions when tested with known anti-M and anti-P sera.

The post-mortem examination revealed marked splenomegaly with some hepatomegaly: focal necrosis of the liver with a fine cirrhosis of portal distribution. The spleen showed marked siderosis, very numerous histiocytes, and absence of lymphoid follicles, the whole gland being intensely engorged with blood. A diagnosis of haemolytic anaemia secondary to Banti's disease was made.

Summary and Conclusions

The patient, who was transfused in the course of a month with blood from 24 different donors, developed a rare antibody (anti-E), a very rare antibody (anti-M, active at 37° C.), and a specific cold antibody (anti-P).

The blood from all of these donors was "compatible" with respect to the A, B, O, and D blood antigens.

As routine methods of cross-matching are inadequate to detect the rarer antibodies it is advisable that special techniques should be employed in those cases in which repeated transfusions are likely to be required.

The details of the laboratory investigations given in this paper clearly show that the search for and identification of such antibodies can be successfully undertaken only in those laboratories where a panel of rare cells, samples of rare antibodies, and specially trained staff are available.

Systematic search for rare antibodies in patients who have received many transfusions of so-called compatible blood will reveal that these antibodies are of much more frequent occurrence than is generally believed.

We are greatly indebted to Drs. R. J. Twort and A. J. Bailey for the clinical notes and post-mortem findings in this case, and to Mr. I. Dunsford for guidance in the serological investigations.

A circular from the Ministry of Health to hospital boards and committees discusses the conditions in which hospital employees aged 15 to 18 should work. The following are among the duties in which they should not be employed: handling drugs or nursing sick people; working in operating theatres or mortuaries; working in homes for the dying, cancer hospitals, or V.D. hospitals or departments. They should not work in the wards of mental hospitals or mental deficiency institutions or in the x-ray, physical-medicine, or out-patient departments of mental hospitals, isolation hospitals, or tuberculosis sanatoria. Young persons under the age of 17 should not be employed in the x-ray, physical-medicine, or out-patient departments of maternity hospitals or in hospital reception rooms, nor should they be attached to wards in any type of hospital. Among the duties considered suitable for young persons the circular lists help with filing, care of linen, attending to the telephone, and clerical work. Hospital authorities should arrange for young persons employed by them to attend part-time courses of further education for the equivalent of one day a week, and this time should be paid for at the usual rates.