giving repeated doses of dimethyl ether of D-tubocurarine iodide by the same route.

We have based the dosage of the dimethyl ether of D-tubocurarine iodide on body weight; it has initially been of the order of 1 mg. per stone (6.35 kg.). We feel that it is safer to dose a patient adequately than to underdose him with any curarizing agent, to lessen the tendency to bronchospasm. Incremental doses of the dimethyl ether of D-tubocurarine iodide are of the order of 3 mg., and their administration is guided by the degree of diaphragmatic activity.

We were impressed by one striking improvement in our results on changing to the new compound—namely, the extreme ease with which a lung or lobe which had been allowed to collapse could be reinflated. Using D-tubocurarine chloride we had always been able to reinflate a lung or lobe, but quite firm pressure on the bag was often required, more particularly in the later stages of the operation. Using dimethyl ether of D-tubocurarine iodide, however, we found that the pressure required to reinflate the lung was much less and remained so throughout the operation. What is the explanation? Is it that when using D-tubocurarine chloride an excess of circulating histamine, with its tendency to increase bronchial muscle tone, is released as the operation proceeds and trauma increases, and that with the use of dimethyl ether of D-tubocurarine iodide there is less histamine release and consequently less stimulation of the tone of the bronchial tree?

There was also a definite change in the blood pressure picture. The fall in systolic blood pressure with an accompanying rise in diastolic pressure previously noticed did not seem to occur to the same extent, or, if it did, was manifest only at a much later stage than with D-tubocurarine chloride.

Conclusions

Owing to the absence, through illness, of one of us (H. B. W.), this communication is not as complete as we would desire and should be regarded as an interim report. With that limitation, our conclusions from using the dimethyl ether of D-tubocurarine iodide as the curarizing agent in 100 cases of major intrathoracic procedures, including pneumonectomies, lobectomies, and mediastinal tumours, are as follows:

1. Dimethyl ether of D-tubocurarine iodide seems to be 2 to 2½ times more potent than D-tubocurarine chloride.
2. The duration of action on diaphragmatic activity is a little shorter than with D-tubocurarine chloride and subsequent doses are required at intervals of 20 to 25 minutes.
3. Clinically there seems to be less release of histamine, as is evidenced by (a) maintenance of systolic and diastolic blood pressures, and (b) ease of inflation of collapsed lung tissues, particularly at the later stages of the operation.
4. In all other ways, as would be expected, the two preparations seem similar in action.

Recent work by Pelikan, Sadove, and Unna (1950) on the relative potency of D-tubocurarine chloride and the curare-like drugs gives support to our clinical findings on the relative strengths of D-tubocurarine chloride and the dimethyl ether of D-tubocurarine iodide, as it does to the relative duration of action of these preparations.

Our impressions are that in the dimethyl ether of D-tubocurarine iodide we have a curarizing agent which has all the advantages of D-tubocurarine chloride and which, from our small series of cases, seems to have less tendency towards histamine release. In no case had we any cause for anxiety, either on the table or post-operatively,

which could be referred to the use of dimethyl ether of D-tubocurarine iodide. However, as of passing interest, it may be noted that in one recent thoracotomy, using D-tubocurarine chloride, a frank bronchospasm occurred and was confirmed by bronchoscopy. Fortunately it responded to appropriate measures.

No evidence of delayed detoxication of the dimethyl ether of D-tubocurarine iodide has been noted, nor have we seen any undue reactions to the drug. The possibility of idiosyncrasy to iodides should, however, be borne in mind when administering it.

The dimethyl ether of D-tubocurarine iodide used in this study was provided by Allen and Hanburys Ltd. through the Joint Anaesthetics Committee of the Medical Research Council and the Royal Society of Medicine.

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NINE BLOOD-GROUP ANTIBODIES IN A SINGLE SERUM AFTER MULTIPLE TRANSFUSIONS

BY

J. O. COLLINS, M.D.

RUTH SANGER, B.Sc., Ph.D.

F. H. ALLEN, Jun., M.D.

AND

R. R. RACE, Ph.D., M.R.C.S.

(From the Clinical Laboratory, Waterbury Hospital, Waterbury, Connecticut; the Blood Grouping Laboratory, Children's Medical Center, Boston; and the Medical Research Council Blood Group Research Unit, Lister Institute, London)

The complete case record upon which this paper is based presents many facets that might be explored with interest by specialists in various fields of medicine and biology. One of the most striking features concerns the large number of antibodies this splenectomized patient was able to produce in response to a comparatively small number of transfusions administered at irregular intervals over a period of several years. Briefly the history is as follows.

Case History

Mrs. F. W., a 40-year-old white housewife, was first admitted to the Waterbury Hospital in September, 1933, for delivery. This was her second pregnancy, the first having terminated with toxæmia and premature stillbirth. After normal delivery of a normal child it was discovered that she had a large spleen. This she stated had been present for 12 years. She was re-admitted for splenectomy in March, 1937. Blood studies on three occasions before splenectomy showed no anaemia or other evidence of blood dyscrasia. The spleen weighed 1,100 g., and increased fibrous tissue with many scattered islands of myelopoiesis and erythropoiesis were seen microscopically. Also many megakaryocytes were noted. Splenectomy was followed by mild anaemia, for which two blood transfusions were given without reaction.

During the next 12 years the patient was repeatedly admitted for anaemia and received several transfusions, as indicated in

Table I. She developed a leuco-erythroblastic peripheral blood picture with corrected white counts in the neighbourhood of 100,000. Her liver enlarged to fill most of the abdomen.

TABLE I.—*Blood Transfusions of Patient F. W.*

	No.	Donors (All Group O)	Reaction
March, 1937	1	Rh?	None
	2	Rh?	"
Feb., 1945	3	Rh+ (anti-D)	"
	4	Rh+	"
	5	Rh+	"
	6	Rh+	"
April, 1946	7	Rh+	"
	8	Rh+	"
	9	Rh+	"
	10	Rh+	"
	11	Rh+	Chill and fever, nausea and vomiting
	12	Rh+	Mild haemolytic
	13	R _u r (anti-C-D-E)	None
	14	R _u r	"
Jan., 1949	15	R _u r	"
	16	R _u r	"
	17	R _u r	"
	18	R _u r	"
	19	R _u r	Mild haemolytic
July, 1949	20	R _u r	Mild febrile

X-ray therapy over her liver resulted in prompt reduction of both red blood cell count and white count. In 1946 attempts to obtain a satisfactory cellular bone marrow by aspirations of the sternum, the right iliac crest, and a spinous process all failed. Skeletal radiographs revealed increased density of many bones with destruction of the normal trabecular pattern. A diagnosis of osteosclerosis and extramedullary haematopoiesis was made. She developed a bleeding peptic ulcer in 1948 and thereafter received a number of additional transfusions. Her transfusion history is set forth in Table I.

Thus it is seen that in a series of 20 transfusions reactions followed Nos. 11, 12, 19, and 20. It was found in 1949 that the patient was Rh_i (CDe) and was sensitized to E. It was also found that her serum agglutinated some Rh_i bloods, so donors of type R_ur (cDe/cde) were chosen. When one of these produced a mild haemolytic reaction, samples of serum and later of whole blood were sent to Boston for more complete studies.

Serological Details

Nine blood-group antibodies were identified in the serum of this patient (Table II). Since four of the antibodies had each been reported on fewer than eight previous occasions

TABLE II.—*The Blood Groups of Patient F. W. and the Antibodies in Her Serum*

System	Phenotype	Genotype	Antibodies in the Serum
ABO	O	OO	Anti-A and anti-B
MNS	MsMs	MsMs	Anti-N and anti-S
Rh	..	CDe/cde or CDe/cDe	Anti-E and anti-C ^w
Kell	..	kk	Anti-K
Lewis* .. .	Le(a- b-)	?Le ^c Le ^c	Anti-Le ^a and anti-Le ^b
P	P-	pp	—
Lutheran* ..	Lu(a-)	LubLub	—

* For notation see Andresen *et al.* (1949).

it is clear that she showed extraordinary powers of antibody production. Such versatility has only once before been recognized, when seven blood-group antibodies were found in the serum of a woman suffering from lupus erythematosus diffusus who had been transfused on a number of occasions (Callender and Race, 1946).

Circumstances caused the serum (F. W.) to be investigated before the full groups of the patient's red cells were determined. This resulted in a situation, probably unique, in which it was possible to tell from the serum the ABO, MNS, Lewis, and Kell groups of the patient and to make a very good guess at the Rh group.

The antibodies are all to be found together in one sample of serum. Five of the antibodies are very rare ones, for

at the time of their recognition in this serum anti-C^w had been found on four previous occasions, anti-S on five, anti-K on five, anti-N on seven, and anti-Le^b on about ten.

In view of the results of agglutination tests in saline at different temperatures, tests in albumin, and indirect anti-globulin tests, it seems certain that the anti-E and anti-K were immune antibodies. The anti-S, anti-C^w, and anti-Le^a all reacted more strongly at room temperature (17° C.) than at 37° C., and all reacted weakly in albumin; by use of the indirect anti-globulin test the anti-S gave a strong positive reaction, and the anti-C^w and anti-Le^a were weakly positive. It seems probable that these three antibodies were immune in origin. The anti-N and anti-Le^b were definitely cold agglutinins and may not have been immune.

Method of Investigation

At the Blood Grouping Laboratory, Boston, tube tests with undiluted serum and saline suspensions of 14 fully grouped and selected blood samples were carried out at 4° C., 17° C., and at 37° C. Tests against the same cells in albumin were also done at 37° C. The results of the albumin tests most clearly showed that anti-E was present. The serum was then tested against 10 samples of cde/cde in saline; at 17° C. all but one of the samples were agglutinated, the one sample being the only S negative in the 10. This made it almost certain that anti-S was present, and this was later proved to be so. The serum was then tested against cells lacking the antigens E and S, and the indirect globulin method showed that anti-K was present.

It was realized that there were still more antibodies present, but the supply of serum was exhausted. About three months later a second sample of serum was obtained, which was dried and sent to England for further investigation.

The presence in the reconstituted serum of anti-E, anti-S, and anti-K was confirmed. The serum was then tested at 17° C. against eight saline suspensions of blood lacking the antigens E, S, and K, and three of the samples were agglutinated: two of these were Le(a+), the third was Le(a-) but contained the antigen C^w. Indirect anti-globulin tests were weakly positive with the two Le(a+) samples and negative with the C^w sample. This suggested that these three reactions were due to two antibodies—probably anti-Le^a and anti-C^w. Cross absorptions and tests on selected samples of blood confirmed this. Re-inspection of the Boston protocols showed that the anti-C^w had at that period been giving a weakly positive anti-globulin reaction.

Tests carried out on a tile resting on ice showed that anti-N was present. The cells used were all Le(a-), E-, C^w-, K-, and S-. The anti-N reaction reached its maximum in 20 minutes.

Next 15 samples of cells which were Le(a-), E-, C^w-, K-, S-, and N- were tested, again on an iced tile, in a moist atmosphere: positive reactions were long-delayed, but were definite after 40 minutes and showed the presence of anti-Le^b. By other methods no positive reactions with the 15 cell samples could be detected.

Discussion

The recognition of such predisposition to make blood-group antibodies as that shown by the patient with osteosclerosis and extramedullary haematopoiesis, and by the patient previously described suffering from lupus erythematosus diffusus, raises a number of questions which cannot at present be answered. Were these two patients born

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.