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Research Article

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PART II. THE EFFECTS OF SPLENECTOMY IN THALASSEMIA MAJOR WITH AN ASSOCIATED ACQUIRED HEMOLYTIC ANEMIA

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PART I

AN EXTRACORPUSCULAR DEFECT IN THALASSEMIA MAJOR

Normal red blood cells, when transfused to patients with thalassemia, have been shown to have a normal life span (1–4). It has also been demonstrated that transfused erythrocytes from donors with thalassemia major rapidly disappear from the circulation of normal recipients (2, 4). This evidence supports the contention that the hemolytic component of the anemia in thalassemia is due to an intracorpuscular defect, as is the case in congenital spherocytic anemia (5) and in sickle cell anemia (6).

We have had the opportunity to observe repeatedly approximately 20 children with thalassemia major during the past five years. All of these subjects have required blood transfusions to alleviate such recurring symptoms as listlessness, irritability, anorexia, fatigue, palpitations, weakness or dizziness. It was evident after review of the transfusion records that many of these children were receiving 500 cc. supplements of blood, or the equivalent in packed erythrocytes, at very short intervals, *i.e.*, every ten days or every 25 days. Since (a) prior to transfusion, hemoglobin and hematocrit levels in any one patient were fairly constant (and definitely not rising over a period of several years), and since (b) there was no external loss of blood, it seemed logical to assume that the transfused blood was not surviving the expected normal span of 120 days. Other possible explanations were that each transfusion caused a decrease in endogenous blood formation or that both of these mechanisms were involved. It was the purpose of this study to test the first assumption by measuring the survival of normal fresh blood in seven recipients with thalassemia major.

METHODS

The survival of transfused erythrocytes was determined by the method of DeGowin, Sheets, and Hamilton (7). Blood from normal type O donors was collected aseptically in vacuum bottles containing "acid-citrate-dextrose" solution and was transfused to the test subjects within 36 hours. Compatibility of donor cells was assured after cross-matching by the use of the indirect Coombs test. Five of the recipients were type A and one was type AB. Anti-A sera were used in dilutions producing minimal inagglutinable cell counts. Sera capable of yielding baseline counts below 25,000 cells per cu. mm. were used. Aliquots of the selected sample of serum were kept in the deep freeze unit and were used for successive measurements during each erythrocyte life span study. One patient whose blood was type O, MN, was transfused with O, N cells. Lederle's anti-M serum was used for agglutination.

The reliability of this method in our hands was demonstrated in normals and in patients with a variety of disorders other than acquired hemolytic anemia. Survival of transfused erythrocytes for 100 to 130 days was found in agreement with results reported by others (7–10).

MATERIAL

The seven patients in this series were examples of severe thalassemia major. All were of Sicilian descent and were first noted to be anemic in infancy, when blood examinations demonstrated red blood cell abnormalities such as microcytosis, hypochromia, basophilic stippling, normoblastosis, target cells, marked anisocytosis and poikilocytosis with many small fragmented forms. The erythrocytes showed increased resistance to hypotonic saline solutions. Examination of bone marrow aspirations

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TABLE I

Hematologic observations in thalassemia major before splenectomy

Case no.	Age *yrs.*	Sex	Average pre-transfusion Hb. level *Gm. %*	Average interval between 500 cc. transfusion *days*	Calculated* whole blood requirements *cc./day*	Measured red cell survival *days*	Half life of transfused red cells *days*
1. P. T.	5½	M	7.0	19.4	26.6	13	7
2. F. L.	8	F	6.0	21.4	23.3	33	8
3. C. A.	7	F	6.5	25.1	19.7	22	8½
4. L. I.	16	M	6.0	10.7	46.6	55	5½
5. D. C.	6	F	7.0	15	31	96	35
6. R. R.	9	M	7.0	19.6	23.4	10	5
7. J. C.	7	M	7.0	18	23.4	26	9

* These figures were calculated by dividing the total volume of whole blood received by transfusion during the previous one to two years by the number of days during this period.

revealed intense myeloid activity with normoblastic hyperplasia. Iron deficiency was excluded by the finding of high serum iron levels and by the failure of the anemia to respond to the administration of preparations containing iron. Splenomegaly was prominent along with moderate hepatomegaly. Each child possessed the mongoloid facial appearance so often noted in this disease (11), in addition to the characteristic roentgenographic abnormalities in the bones (12). Detailed hematologic, genetic and roentgenologic studies of these patients will be reported elsewhere.

RESULTS AND DISCUSSION

The circulating "life span" of normal red blood cells transfused to these patients was markedly shortened in all but one instance (see Table I and Figures 1–5). The "half life" of the transfused erythrocytes ranged from five to nine days in six of the patients. In Case 5, where the total survival time of the transfused red cells approximated 96 days, the "half life" of these cells was only 35 days. The rate of elimination of the cells in this patient, like those of Cases 2 and 4 (see Figure 2 and 4), is such that it appears curvilinear when charted. This has been described by Brown, Hayward, Powell, and Witts as characteristic of a random destructive mechanism which is independent of the age of the cells (13). The shortened sur-

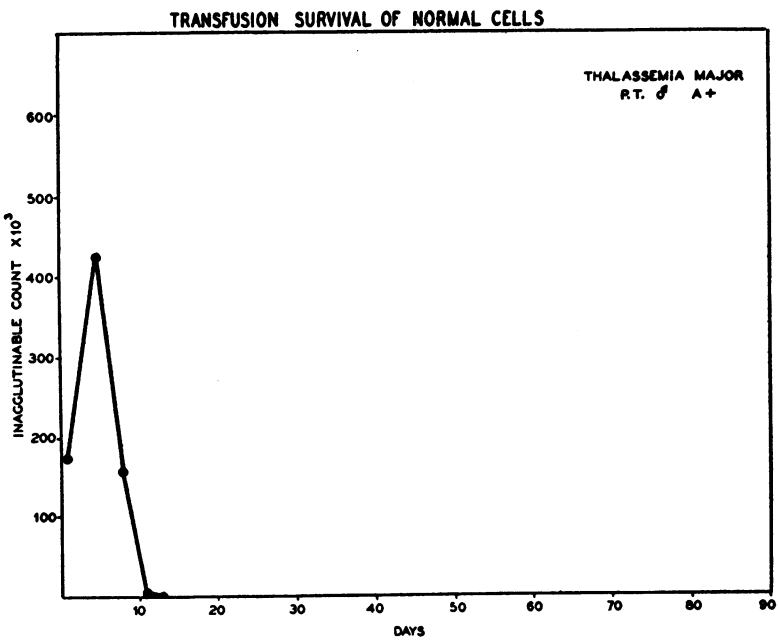

FIG. 1. CASE 1

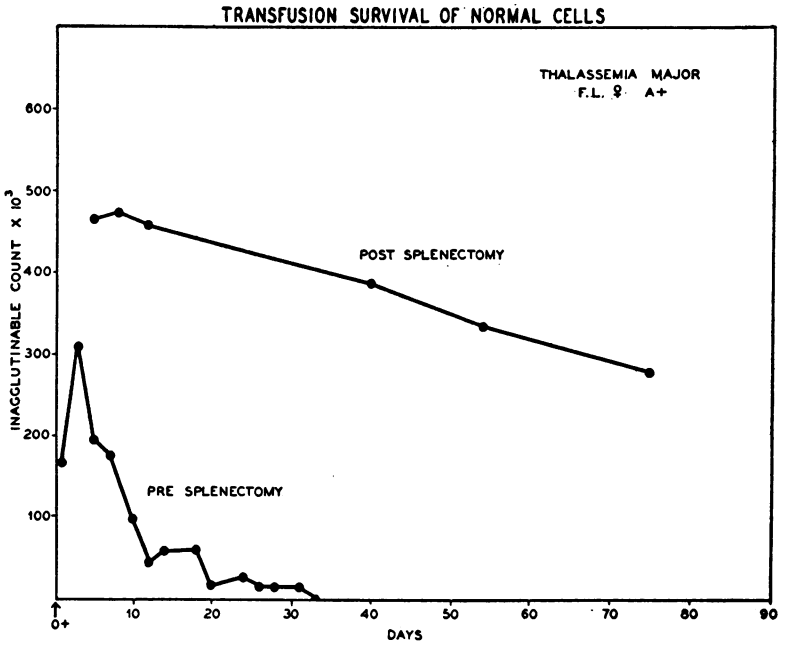

FIG. 2. CASE 2

vival of transfused red cells indicates that in all of these patients an extracorpuscular type of hemolytic anemia exists in addition to any other mechanisms operative in thalassemia major.

In four previously reported studies (1–4) on a total of seven separate patients with thalassemia the survival of normal transfused erythrocytes was found to be normal. Sufficient data are available in five of these studies to demonstrate differences in case material which may explain the discrepancy between their results and ours. Four of the previously reported patients were below two years of

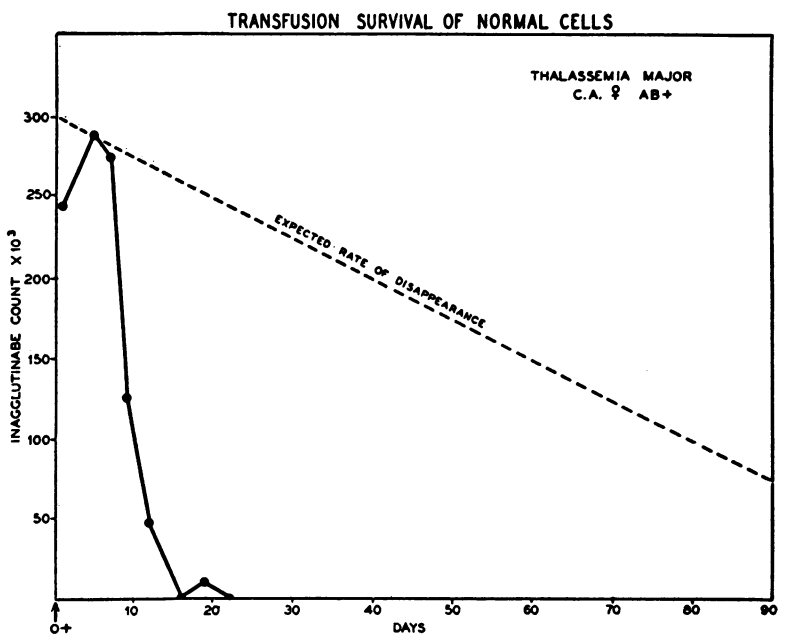

FIG. 3. CASE 3

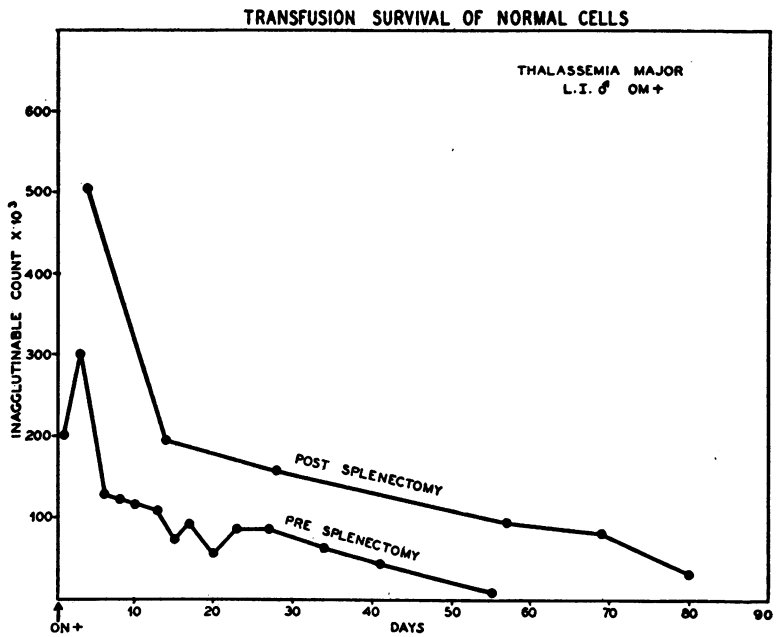

FIG. 4. CASE 4

age, and one who was eight years old had never required any blood transfusions (see Table II). The patients included in the present study were between five and 16 years old. Their records indicate that increased demands for transfusions did not become evident before the age of three.

It may be that the development of an extracorpuscular hemolytic anemia in thalassemia is dependent on an immunologic mechanism related to the large number of transfusions received by some of these patients over a period of several years. Circulating antibodies have not been demonstrated

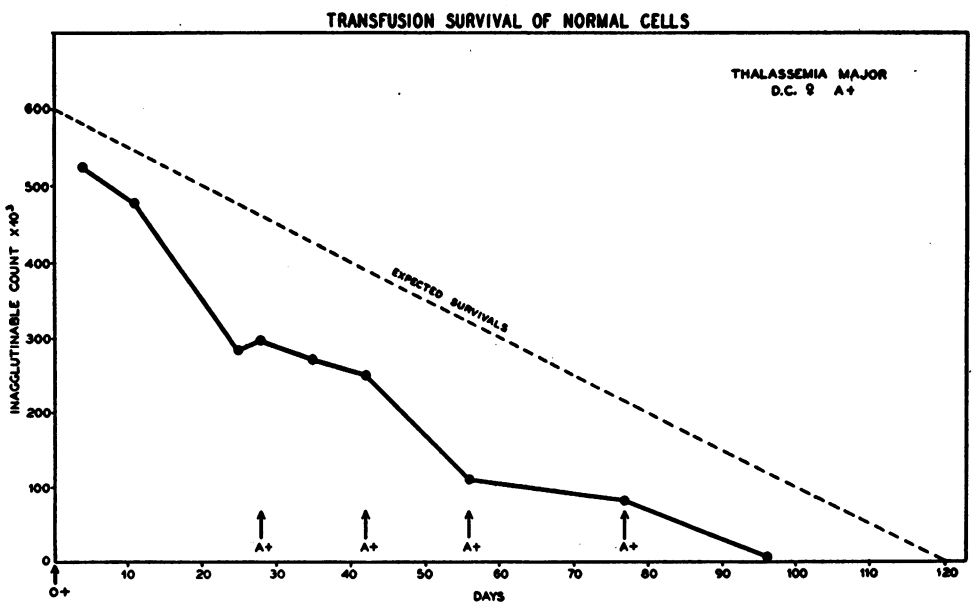

FIG. 5. CASE 5

TABLE II

Previously reported cases of thalassemia with normal erythrocyte survival of transfused cells

Case no.	Age	Frequency of transfusions	Observer
1	8 yrs.	Never transfused	Kaplan and Zuelzer (2)
2	2 yrs.	56–84 days	Kaplan and Zuelzer
3	7 wks.	Never transfused	Hamilton, Sheets, and DeGowin (1)
4	9 mos.	?	Frontali and Stegagno (4)
5	18 mos.	?	Frontali and Stegagno
6	?	?	Neber and Dameshek (3)
7	?	?	Neber and Dameshek

in our patients by the use of trypsin treated red cells (14) or by the indirect Coombs test (15). Further immunologic studies are in progress.

The persistent marked splenomegaly increasing with age suggested the possibility that the extracorpuscular hemolytic component was related to the development of an abnormal function of the spleen in these patients. In Part II of this report the role of the spleen will be assessed.

SUMMARY

1. Seven children with thalassemia major who required frequent blood transfusions in order to maintain hemoglobin levels above 6 to 7 Gm. per cent were studied with a modification of the Ashby test for survival time of transfused normal erythrocytes.

2. In each instance a shortened survival of the transfused erythrocytes was noted. This finding demonstrates that an extracorpuscular hemolytic anemia was present in these patients in addition to any other mechanisms operative in the pathogenesis of the anemia in thalassemia major.

PART II

THE EFFECTS OF SPLENECTOMY IN THALASSEMIA MAJOR WITH AN ASSOCIATED ACQUIRED HEMOLYTIC ANEMIA

Surgical removal of the spleen has been demonstrated repeatedly to be helpful in certain patients with hemolytic anemia (16–19) and curative in others (19, 20). How this procedure exerts its beneficial effect has been the subject of much investigation and speculation but a definitive explanation of this mechanism has not yet been supplied. Furthermore, in the presence of the extracorpuscular type of hemolytic anemias there are no reliable criteria available with which one can predict whether splenectomy will be of benefit.

Because of the clear evidence of the existence of an extracorpuscular hemolytic anemia in the seven patients with thalassemia major described above, splenectomy was performed in each instance. It is the purpose of this report to assess the value of the the operation in these selected cases.

RESULTS

The data on transfusion requirements of five patients who have been observed for longer than six months following splenectomy are summarized in Table III. A comparison of rates of blood replacement can also be seen in Figure 6. Transfusion requirements fell significantly during the period of observation in four instances, *i.e.*, 19 per cent, 21.3 per cent, 28.3 per cent and 36 per cent of preoperative figures. The fifth patient (Case 4) was not benefited. His requirements for blood after splenectomy were 108 per cent of preoperative levels. In the first four patients the intervals between transfusions of 500 cc. of whole blood increased from a range of 15 to 25 days before operation to a range of 57 to 119 days after splenectomy.

Actual measurements of the survival of the transfused blood were made, using the previously described modification of Ashby's test in two instances after splenectomy. As demonstrated in Figure 2, the rate of elimination of transfused erythrocytes after splenectomy was normal, in sharp contrast to the markedly shortened preoperative cell survival. This patient (Case 2) was greatly improved following splenectomy in that her transfusion interval increased from 21 days to 75.0 days.

In Case 4 (Figure 4) the half life of transfused red cells is seen to be only slightly increased, *i.e.*,

TABLE III

Post splenectomy—calculated whole blood requirements

Case no.	Average interval between 500 cc. transfusions	Calculated whole blood requirements	Fraction of pre-operative transfusion rates	Period of observation post-operatively
	days	*cc./day*	%	*months*
1. P. T.	57.7	8.6	36.0	12
2. F. L.	75.0	6.6	28.3	12
3. C. A.	119.0	4.2	21.3	12
4. L. I.	9.8	50.5	108.	9
5. D. C.	84.0	5.9	19.0	6

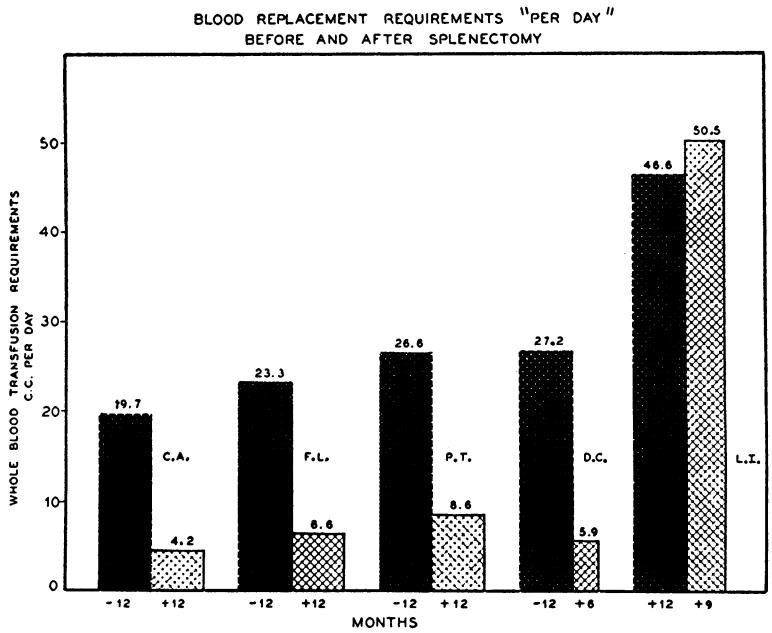

FIG. 6

14 days as compared to five and one half days. This was the single patient studied whose transfusion requirements were not improved by splenectomy.

DISCUSSION

Splenectomy has been performed in thalassemia major many times in the past. Some (21, 22) have recommended the operation for its beneficial hematological effects. Most observers feel that splenectomy is productive of little clinical improvement apart from the mechanical relief gained by removal of a large and heavy organ from the persistently distended abdomen (23, 24).

The present study indicates that in a select group of patients, splenectomy is of value in most instances. The selection in this series depended on the findings of an extracorpuscular type of hemolytic anemia. This was demonstrated by red cell survival measurements. It is our impression that the chronic pronounced splenomegaly, persisting for at least three years, was instrumental in causing a rapid destruction of normal transfused blood cells. That the spleen may not be the only organ involved in this pathological capacity is suggested by a lack of improvement in Case 4 following operation. It is of interest that in the entire series this patient had the greatest degree of hepatic enlargement.

An extracorpuscular hemolytic anemia can be predicted with relative confidence when: 1) Transfusions are required to maintain adequate hemoglobin levels at intervals shorter than the normal survival of transfused blood; and when 2) there is no other cause for a rapid fall in circulating hemoglobin concentration such as bleeding or the expansion of the intravascular space, or a temporary decrease in endogenous erythrocyte production. That variations in endogenous erythrocyte production do occur in thalassemia has been demonstrated (1). Just how important this factor is in affecting the levels of circulating hemoglobin in thalassemia awaits further study. It is tempting to postulate that the discrepancies between the measured red cell survival and the required interval between blood transfusions are related to fluctuations in endogenous erythrocyte production. This is suggested in Case 5 (Figure 5). A transfused red cell survival of 96 days with a "half life" of 35 days was found. In spite of this however, administration of 500 cc. of blood was required at intervals of 15 days in order to keep the circulating hemoglobin concentration above 7 Gm. per cent.

The number of circulating erythrocytes present in the body at any specific moment is a function of four variables: 1) The amount of blood transfused; 2) the rate of destruction of these cells; 3) the rate of endogenous production of erythrocytes; and 4) the rate of destruction of these endogenously produced cells. In this study we have data on the first two variables. Knowledge of the dynamics of factors 3) and 4) is necessary for a more complete understanding of the mechanism of the anemia in these patients.

SUMMARY

1. Five patients with thalassemia major in whom an extracorpuscular type of hemolytic anemia was demonstrated, were subjected to surgical removal of their spleens.

2. The requirements of four of these patients for supportive blood transfusions were markedly reduced. One patient was unchanged clinically.

3. The survival of transfused erythrocytes was measured in two instances after splenectomy. In one instance the transfused erythrocytes were found to have a normal life span after operation as compared to a total survival of 33 days before splenectomy. In the second patient who was not benefited by splenectomy there was no significant change in the survival of transfused erythrocytes.

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