Reticulocytopenia in Autoimmune Hemolytic Anemia

By WILLIAM H. CROSBY AND HENRY RAPPAPORT

Reticulocytosis is generally regarded as a characteristic feature of the hemolytic syndrome. The outpouring of young red cells is one of the bone marrow’s reactions to the challenge of anemia, and the number of reticulocytes it produces is often a good measure of the severity of the hemolytic process. When the red cell life span is very short most of the cells in the circulation may be reticulocytes, indicating that few cells live much beyond their reticulum-bearing babyhood. Pernicious anemia is an important exception. The bone marrow in pernicious anemia may produce four or five times the normal number of red cells yet the reticulocyte count is low. Examination of bone marrow from cases of pernicious anemia reveals the reason for this. Vital staining of aspirated marrow demonstrates ample reticulum substance in the cytoplasm of the nucleated erythroid cells. But the cytoplasm matures and the reticulum substance disappears before the nucleus is lost. As a consequence, the young cells issuing from the marrow are not reticulocytes. Another instance of reticulocytopenia in the presence of hemolytic disease occurs during the regenerative or aplastic crises that may strike patients with chronic hemolytic anemia of various kinds. The regenerative crisis was first identified in patients with hereditary spherocytosis, but it also occurs in autoimmune hemolytic anemia. For one reason or another the bone marrow abruptly stops its hematopoietic activity and may actually become aplastic. Then intervenes a period of reticulocytopenia, leukopenia and thrombocytopenia. Because hemolytic disease is already present severe anemia rapidly develops. Fortunately the crisis is usually short lived. In conditions where the bone marrow is obliterated by neoplastic or fibrous tissue hemolytic disease and reticulocytopenia may occur together. With these exceptions there is a general impression that it is unusual to find severe hemolytic disease, erythroid hyperplasia of the marrow and reticulocytopenia simultaneously.

In a survey of cases of autoimmune hemolytic anemia (AHA) from the files of the Armed Forces Institute of Pathology it was noted that a number of patients exhibited, at one time or another, a definite reticulocytopenia in the presence of severe hemolytic anemia with a bone marrow that presented a picture of intense erythroid proliferation. The phenomenon appears to have a sinister significance.

Materials and Methods

The selection and methods of analysis of 57 cases of AHA have been described elsewhere. Reticulocytosis is not arbitrarily defined, but it is considered in relation to the patient’s hemoglobin concentration. Thus with 5 Gm. of hemoglobin a 5 per cent reticulocyte count is considered reticulocytopenic, while with 15 Gm. of hemoglobin the same percentage represents reticulocytosis, because in terms of absolute numbers of reticulocytes it represents three times as many cells. A glance at figure 1 will demonstrate that there is no difficulty in...
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Reticulocytosis and Anemia in Crises of Idiopathic Autoimmune Hemolytic Anemia (34 Cases)

Fig. 1.—Reticulocyte counts versus red cell counts in 34 cases of idiopathic autoimmune hemolytic anemia. A caret on the abscissa is at point representing a hemoglobin concentration of 7 Gm. per 100 ml.

identifying the reticulocytopenic cases. They form a discrete cluster in the lower left corner. In the study of the records of these patients reticulocytosis or reticulocytopenia was not determined on the basis of a single count that might have been wrong. The count selected as representative was one of a series. The figures for comparison of patients with reticulocytosis and those with reticulocytopenia reflect the picture at times of hemolytic crisis. It was endeavored to establish the reticulocyte count when the hemoglobin concentration was lowest.

Results

Of 34 patients with idiopathic AHA, 15 were found to have relative reticulocytopenia at times of hemolytic crisis and only 3 of them survived; of those with severe anemia whose hemoglobin concentration was less than 7 Gm., only one survived (fig. 1). In all of these the marrow was reported to be hyperplastic. The rest of the patients were considered to have a reticulocyte response that was consistent with the severity of their anemia.

Of 16 patients with AHA secondary to lymphoma, 10 had reticulocytopenia. All but two of these 16 patients are dead. This is not a useful correlation because neoplastic disease may affect bone marrow function either directly or indirectly, and because the lymphomas are invariably fatal.

Of 7 patients with AHA secondary to nonmalignant conditions (lupus erythematosus, infectious mononucleosis, sarcoidosis and Felty's syndrome) only two had reticulocytopenia. One was a woman with lupus who died of lupus several years after the crises of AHA had passed. The other was one of three young men with infectious mononucleosis.
In two patients reticulocyte counts were not reported. Four illustrative cases are shown in figures 2–5, demonstrating reticulocytopenia in fatal and nonfatal idiopathic AHA, and in symptomatic AHA associated with benign and malignant diseases.

Fig. 2.—Idiopathic AHA and idiopathic thrombocytopenic purpura (ITP). (AFIP Accession 311839). A 20 year old man, every spring during his eleventh, twelfth and thirteenth years, had had purpura that lasted for three or four months. His final illness began abruptly on July 20, 1950, with nosebleed and dark urine. He was admitted to a station hospital and on August 4 was transferred to an Army general hospital. Physical examination showed light icterus, pallor and a barely palpable spleen. Hemoglobin 7 Gm. WBC 7,000 with 72 per cent neutrophils. Platelets 13,500. Reticulocytes 9 per cent, later fell to 3. Bone marrow was hyperplastic with M:E ratio of 1:1 and many inactive megakaryocytes, a picture consistent with AHA plus ITP. The Coombs test was negative at first, but became positive. It was difficult and then impossible to find compatible donors. After splenectomy on August 10 there was a fleeting remission but he became severely anemic and died August 30. Sections of the spleen showed an extreme degree of erythropagocytosis and also extramedullary hematopoiesis, predominantly erythroid in type. Postmortem examination revealed severe pulmonary edema without evidence of intrinsic cardiac disease. The pulmonary edema is attributed to myocardial insufficiency secondary to severe anoxia incident to the anemia. The liver showed very severe anoxic necrosis and also extramedullary hematopoiesis. Erythropagocytosis was present in lymph nodes and bone marrow. Note that both the reticulocyte count and platelet count were elevated temporarily after splenectomy.
Fig. 3.—Idiopathic AHA. (AFIP Accession 285665). The illness of this 27 year old man had a stormy, febrile onset November 17, 1949. He was transferred from a station hospital to an Army general hospital November 20. He was pale, moderately jaundiced with a palpable spleen. Hemoglobin 3 Gm. WBC 16,200 with 76 per cent neutrophils. Platelets 86,000. Reticulocytes 0.1 per cent. Osmotic fragility normal. Bilirubin 5.7 mg. (4.5 mg. indirect). Plasma hemoglobin 120 mg. Coombs test: slight agglutination. There was marked erythrophagocytosis in the aspirated bone marrow and in smears of the buffy coat. The bone marrow also showed intense erythroid hyperplasia. Until December 1 he had received 19 transfusions and 14 more in the next two weeks. Hemoglobin remained between 5 and 9 Gm. and repeated reticulocyte counts were never above 1 per cent. Splenectomy was done on December 12. He needed no transfusions after that, but the hemoglobin remained low, around 6 or 8 Gm. and his reticulocytes were one per cent or less. The last week in January the reticulocytes began to increase and went to 15 per cent. Then the hemoglobin rose to 10 and then to 12 Gm. but by this time, with the anemia corrected, the reticulocytes again subsided and his marrow became normal. This is the only patient with severe anemia and reticulocytopenia who survives (fig. 1).

DISCUSSION

The number of reticulocytes in the circulating blood depends upon two factors, the number of red cells delivered from the bone marrow with reticulum substance still present and the length of time that the reticulum substance remains in the cells. The life span of reticulocytes as such in the circulation has not been clearly established but it apparently depends upon at least two factors: the amount of reticulum present in the cell when it leaves the bone marrow and the rate at which the reticulum is disposed of. The younger cells have more reticulum. Thus if a cell is released early from the bone marrow it remains a reticulocyte for a
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AUTOIMMUNE HEMOLYTIC ANEMIA WITH INFECTIOUS MONONUCLEOSIS

![Graph showing hemoglobin (HB), white blood cell (WBC), reticulocyte, and other parameters over time.]

Fig. 4.—Symptomatic AHA with infectious mononucleosis (AFIP Accession 587588).
The illness of this 19 year old man began on April 1, 1953, with gradual onset of fever and malaise. Symptoms of anemia developed and on April 8 he passed dark red urine. He was transferred to an Army general hospital on April 11. He demonstrated pallor, light icterus, hepatosplenomegaly and a mild, generalized lymphadenopathy. Hemoglobin 4 Gm. Reticulocytes 2.5. WBC 19,000 with 45 per cent neutrophils and 30 per cent atypical lymphocytes. Bone marrow showed erythroid hyperplasia. Osmotic fragility test showed hemolysis in 0.9 per cent saline (? hemoglobinemia). Heterophile antibody test was positive 1:1800 after adsorption with guinea pig kidney; cold agglutins 1:64; direct Coombs test positive. It was impossible to type and cross match so he was transfused with group O blood without reaction. He was also given 400 mg. of cortisone. This daily dose was rapidly reduced to 100 mg. as improvement occurred. Reticulocyte response was dramatic, rising to 20 per cent as the hemoglobin increased, and as the hemoglobin came to normal levels the reticulocytosis subsided.

Longer time in the blood. The average reticulocyte life span is probably about one to two days in normal adults. In severe hemolytic disease with delivery of very young red cells, the life span of the reticulocytes may be increased.

The percentage of reticulocytes versus absolute number merits consideration.
327642

Fig. 5.—Symptomatic AHA with malignant lymphoma, follicular type (AFIP Accession 327642). Onset of this man's illness occurred in April 1948 with fever, jaundice and anemia. He died in April 1951, age 54, four months after the episode described here following a short period of severe dyspnea and fever with physical manifestations suggesting pulmonary infarction. No autopsy was done. On October 11, 1950, he completed a course of X-ray therapy, 1200 r in air over the spleen. His hemoglobin at this time was 8 Gm. Reticulocytes 1.5 per cent. WBC 6,300 with 70 per cent neutrophils. Platelets 166,000. Coombs test strongly positive. From November 28 until December 28 he was given cortisone by mouth, 100 to 200 mg. per day. The details of this therapy and his response are shown above. Note that the hemoglobin was not increased in spite of the increased reticulocyte count. This suggests that during the period of reticulocytosis the bone marrow was releasing younger red cells but releasing them in no greater numbers.

The upper limit of reticulocytes in normal human blood is generally accepted to be two per cent of the total red cells. When the red cell count is five million per cu.mm., 2 per cent represents an absolute reticulocyte count of 100,000. When the red cells are one million, the same absolute reticulocyte count represents 10 per cent of the total red cells, a degree of reticulocytosis which, in severe anemia, does not represent much of an effort by the bone marrow. It is in fact a relative reticulocytopenia.

The concept of a "reticulocyte pool" in the bone marrow is of help in the interpretation of problems of reticulocytopenia. This concept is based upon the probability that maturing red cells are not usually released by the bone marrow at
the instant they lose their nuclei. The young reticulocytes continue to mature for a variable time, and during the interval before they are called to active duty they provide a reserve of red cells. The usual career in this reticulocyte pool is conceived to be an orderly progression of entering, stopping and departing, and the average time spent there is fairly constant. But any sudden demand for red cells—hemorrhage or hemolysis—draws first upon this pool. In such a case the pool's size is reduced, which is to say, the average time spent in the pool by the reticulocytes is shortened. If the demand is sustained, the pool remains small. The reticulocytes entering the blood are all quite young and even some nucleated red cells may be called up. Reticulocytopenia in hemolytic disease is undoubtedly concerned with the reticulocyte pool: abnormalities of the mechanisms that control its size, injury or destruction of the red cells that comprise it, disturbances of the erythroblasts that supply it with young reticulocytes.

Reticulocytopenia in the presence of severe AHA may be due to several mechanisms other than marrow aplasia or the replacement of marrow by abnormal tissue or an acute "aregenerative crisis.”

1. There may be asynchronous maturation of cytoplasm and nucleus of the erythroblasts, as there is in pernicious anemia. The cells lose the reticulum substance before they lose their nuclei, and the young red cells delivered into the circulating blood are not reticulocytes.

2. Release from the bone marrow of normally maturing erythrocytes may be delayed until the reticulum has disappeared.

3. In acute severe anemia the hypoxia due to lack of hemoglobin may inhibit erythropoiesis even though the marrow remains hyperplastic.

4. Active erythropagocytes in the marrow may destroy maturing erythrocytes in sufficient numbers to prevent their appearing in the blood.

5. Autoimmune disease may damage the proliferating marrow cells as it does the circulating blood cells in such a way as to impede erythropoiesis. In idiopathic thrombocytopenic purpura there is an analogous situation when the antiplatelet antibodies are apparently able to injure the marrow megakaryocytes and prevent their making platelets.

6. The autoimmune mechanism may be selectively active against reticulocytes, destroying them in preference to red cells with no reticulum.

7. Therapeutic agents may inhibit erythropoiesis and the delivery of reticulocytes.

Any of these mechanisms, by impeding the supply of red cells from the marrow, may contribute to the severity of a patient's anemia. Whatever its cause may be, reticulocytopenia as a complication of AHA seemed to carry a serious prognostic significance in the series of cases on which this report is based. Of the patients in the idiopathic group with reticulocytopenia and severe anemia, 7 Gm. or less of hemoglobin, 10 of 11 died (91%); of those with severe anemia, 7 Gm. or less, but an adequate reticulocyte response, 5 of 17 died (29%). It is not intended to imply that reticulocytopenia was a cause of death, although it may have contributed to the anemia. It seems reasonable to suggest that the patients with reticulocytopenia may have had a more severe autoimmune disease or were otherwise more seriously ill than those whose marrow responded to anemia with a high-grade reticulocytosis.
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SUMMARY

1. In a series of 57 cases of autoimmune hemolytic anemia it was found that 25 or 44 per cent had a relative reticulocytopenia at times of hemolytic crisis. The mortality rate in this group was significantly higher than in those who showed a reticulocyte response consistent with the severity of anemia. Special significance is given to the high mortality rate among the patients with idiopathic AHA.

2. Four illustrative cases are presented.

3. There is a discussion of mechanisms that may be responsible for reticulocytopenia in the presence of severe anemia and erythroid hyperplasia of the marrow.

SUMMARIO IN INTERLINGUA

1. In un serie de 57 casos de autoimmmun anemia hemolytic il esseva constatat que 25 (44 pro cento) habeva un relative reticulocytopenia a tempores de crise hemolytic. Le mortalitate in iste gruppo esseva significativemente plus alte que in le gruppo de casos in que le responsa reticulocytic correspondeva al severitate del anemia. Un signification special es attribuite al alte mortalitate inter patientes con idiopathic anemia hemolytic autoimmun.

2. Es presentate quatro casos illustrative.

3. Es discutite le mechanismos que es possibilemente responsabile pro reticulocytopenia in le presentia de sever anemia e hyperplasia erythroide del medulla.

REFERENCES

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