ACUTE TRANSITORY ERYTHROBLASTOPENIA IN KWASHIORKOR

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Acute transitory erythroblastopenia (A.T.E.), or aplastic crisis, was first reported in kwashiorkor patients by Kho Lien-Keng[1] at Djarkarta in 1957. We first noted it in 1960, and soon realized that it was not an uncommon finding in such patients.[2] Walt *et al.*[3] independently noted similar findings in Durban, while Foy *et al.*[4] recorded a red-cell aplasia (seemingly of different type) in kwashiorkor cases in Kenya. Some years earlier, an acute transitory cessation of red-cell production had been noted in association with congenital haemolytic jaundice by Owren[5] in 1948, and with various infections and toxic and allergic conditions by Gasser[6] in 1949. Later, erythroblastopenia was described in association with sickle-cell anaemia,[7, 8] haemoglobin-E thalassaemia[9] and typhoid.[10]

The crisis is characterized by sudden reticulocytopenia and absence of polychromatic cells in the blood, which continues for about 10 days. The bone marrow shows almost complete absence of the erythroid series. Occasional proerythroblasts are found and, in most cases, giant cells resembling pronormoblasts or primitive reticular elements, which have been called giant proerythroblasts.

This paper records the haematological findings in 102 cases of acute transitory erythroblastopenia studied over the past 3 years at King Edward VIII and Clairwood Hospitals, Durban. The purpose of the study was to observe the changes in the blood and bone marrow occurring just before, during, and after the aplastic crisis. The clinical findings will be described in a subsequent paper.

MATERIAL, INVESTIGATIONS AND METHODS

One hundred and two kwashiorkor patients conforming to the criteria of Brock *et al.*[11] and in whom acute transitory erythroblastopenia was found, were studied from 1960 till 1963.

Blood was obtained from the internal or external jugular vein in all patients. The following investigations were performed in the majority of patients.

1. Haematological investigation included a weekly full blood count, reticulocyte count, and platelet count, and in most cases a weekly bone-marrow aspiration. Daily haemoglobin and reticulocyte counts were performed by skin puncture. In some patients, more frequent white blood-cell counts, platelet counts and differential smears were done. In 32 cases serial marrow aspirations were performed in order that the bone-marrow changes might be studied just before, during, and after the aplastic crisis.

Standard haematological methods were used. The platelet count was done by a formol-citrate diluent method using a 1:100 dilution of venous blood, and was controlled by examining direct blood smears (normal = 150,000 - 450,000 per cu.mm.). The marrow was aspirated by iliac puncture and stained with May-Grünwald Giemsa.

2. Serum iron was estimated weekly by the method described by Peters *et al.*[12] and marrow-haemosiderin grading as described by Rath and Finch.[13]

RESULTS

1. Haematological Findings

Acute transitory erythroblastopenia occurred both in normoblastic and in megaloblastic marrows. The sequence of events consisted of a degenerative phase, an aplastic phase, and a recovery phase.

Degenerative Phase

Marrow aspirations from 27 cases were studied during the degenerative phase. Of these marrows 16 were megaloblastic and 11 were normoblastic. At the onset, there was a sudden and rapid degeneration of the erythroid precursors in the bone marrow. This change apparently took place over a period of 24 - 72 hours. At first degeneration occurred among the polychromatic and basophilic cells, but an equally severe degeneration of the earlier members of the erythroid series soon followed. Many of the cells undergoing degeneration showed lustreless nuclei with irregular clumping of the chromatin (karyolysis), while other cells showed homogeneous nuclei (Fig. 1). The cytoplasm of these cells was often fragmented and, in some of the late normoblasts, basophilic stippling was noted. It will be seen from a study of Table I that the rapid degeneration is confirmed by the myeloid:erythroid ratios of the serial marrow aspirations (Figs. 2A, 2B and 2C).

In some normoblastic marrows, during this period of degeneration a few multinucleated and larger cells appeared, and at this stage it was sometimes difficult to decide whether the marrows were not, in fact, partially megaloblastic. The larger cells resembled megaloblasts because of the large size of the nucleus and irregular staining of the nuclear chromatin.

In addition, giant proerythroblasts (25 - 50μ) and giant reticulum cells (40 - 60μ) became apparent, and were

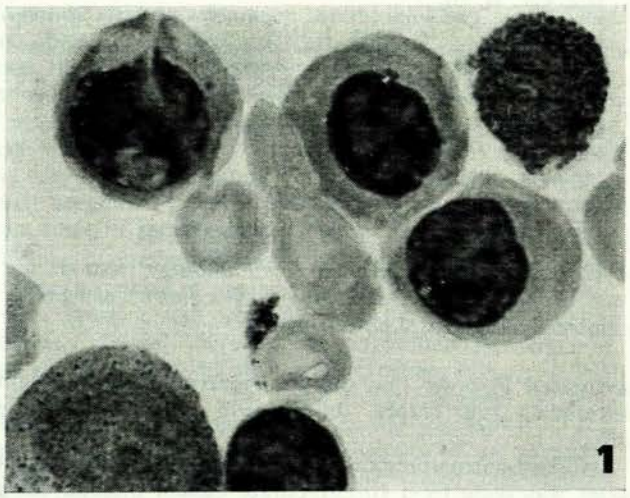

Fig. 1. Erythroid cells in the degenerative phase.

easily found under low-power microscopic examination. The former resembled the pronormoblast (Fig. 3), but was sometimes similar to a promegaloblast, having a more 'open' nuclear structure (Fig. 4). The latter type was

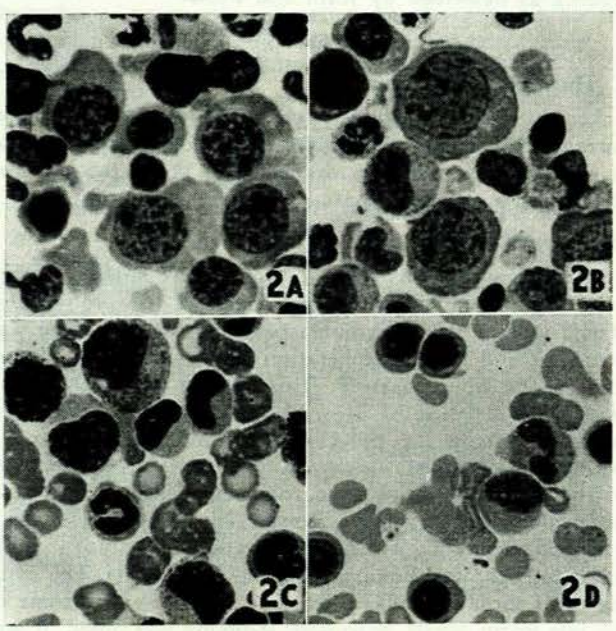

Fig. 2. Serial marrow aspirations, case 57, group D (× 600).

A. Bone marrow on 5 December 1962. Early degenerative phase, showing cells resembling megaloblasts.
B. Bone marrow on 6 December. Degenerative phase, showing large degenerating erythroid cells, not seen in Fig. 2A.
C. Bone marrow on 7 December. Aplastic phase, showing absence of erythroid cells. Two nuclear remains can be seen.
D. Bone marrow on 20 December. Recovery phase, showing normoblastic hyperplasia.

probably a giant proerythroblast showing degenerative changes (karyolysis). The cytoplasm of the cell that resembled the pronormoblast was dark blue and sometimes contained vacuoles, while the cytoplasm of the other type was light blue in colour. The nucleoli of all the giant cells were large, and in some a very large single nucleolus was

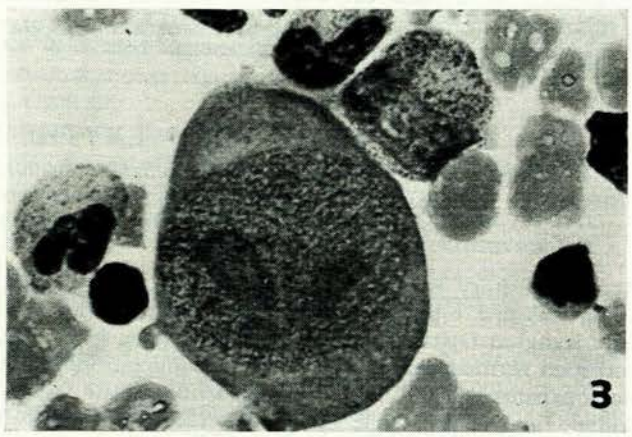

Fig. 3. Giant proerythroblast (37·5μ), with large nucleus (25μ) containing nucleoli, and dark blue cytoplasm.

present. The giant proerythroblasts were more numerous at the beginning of the aplastic crisis, and seemed to decrease in number during the succeeding period. It appeared from serial marrow studies that many of these cells degenerated (Table I).

During the degenerative phase the peripheral blood quite often showed a reticulocytosis (Fig. 5), but this was followed, sometimes very suddenly, by a reticulocytopenia. During this early period it was not possible to diagnose the aplastic crisis from the blood findings.

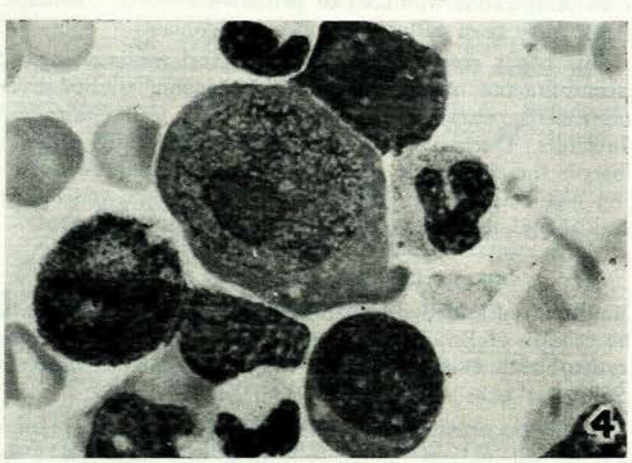

Fig. 4. Giant proerythroblast (25μ), with the nucleus (20μ) showing 'open' chromatin network and containing a large single nucleolus. The cytoplasm stained light blue.

Aplastic Phase

Marrow aspirations were studied from 85 cases in the aplastic phase. This stage was merely a continuation of the degenerative phase, with the erythroid cells almost

TABLE I. (CASE 1, GROUP D), SHOWING MYELOID : ERYTHROID RATIOS, THE DIFFERENTIAL DISTRIBUTION OF THE ERYTHROID CELLS IN SERIAL MARROW ASPIRATIONS, AND THE HAEMOGLOBIN AND RETICULOCYTE COUNT OF THE BLOOD

	1†	2	3	4	5	7	9	12	13	14	16	17	18	20
Blood:														
Haemoglobin G. per 100 ml. ..	6·0	5·3	5·3	5·3	5·1	4·6	4·0	*	8·4	7·9	8·4		8·2	9·3
Reticulocytes %	7·2	10·2	7·6	1·2	0·6	0·0	0·0		2·2	4·6	9·4	14·8	5·8	4·0
						Reticulocytopenia								
Marrow aspirations:														
M : E ratio (1,000 cells)	3:1	9:1	50:1			1,000:1					1·3:1			
Differential distribution of the erythroid cells: %														
Reticulum cell	0·5 (0%)‡	0·0 (0%)‡	0§			0					0·0 (0%)‡			
Giant proerythroblast	0·5 (0%)	8·0 (81%)	0			0					0·0 (0%)			
Proerythroblast	1·0 (0%)	14·0 (89%)	1 (100%)‡			0					0·4 (0%)			
Basophilic erythroblast	13·0 (85%)	23·5 (98%)	1 (100%)			0					2·4 (0%)			
Polychromatic erythroblast ..	69·0 (61%)	34·5 (100%)	2 (100%)			0					14·6 (0%)			
Orthochromatic erythroblast ..	13·5 (4%)	19·5 (95%)	16 (100%)			1 (100)‡					82·4 (0%)			
Multinucleated cells	2·5	0·5	0			0								
Remnant nuclei among 1,000 cells	53	54	92			0					0			
Total % degenerate erythroid cells	54	95	+95			100					0			

*65 ml. of packed cells administered on the 12th day from recognition of aplastic crisis.
†Day when onset of aplastic crisis was recognized—11th day in hospital. ‡Percentage of cells showing degenerate changes.
§Only 20 erythroid series in 1,000 cells counted.

completely disappearing from the marrow. The erythroid cells became shrunken and distorted, their cytoplasm disintegrated, and free nuclear remains increased in number. During the crisis an occasional giant proerythroblast and a few proerythroblasts were present. The number of giant proerythroblasts varied from 1:1,000 to 6:1,000. In 20 cases giant proerythroblasts were not observed, and in these instances proerythroblasts in varying numbers were noted. In 45 cases large reticulum cells (Fig. 6) were seen, and in 4 cases there was an increase in the number of plasma cells. Toxic granulation and a shift to the left was present in the majority of cases, while megakaryocytes were usually observed in normal or increased numbers. In some cases a few abnormal mitoses were noted (Fig. 7).

During the aplastic phase complete absence of reticulocytes was quite common and lasted from 5 to 17 days, most commonly 7 - 10 days. At the commencement of the aplastic crisis the haemoglobin varied from 3·5 to 12 G. per 100 ml. and during the crisis the haemoglobin dropped, in most cases by 0·5 - 1·5 G. per 100 ml., but by as much as 4 G. per 100 ml. in one case. The peripheral blood during this period showed no polychromatic cells. There was no accompanying leukopenia, and thrombocytopenia was noted in only 4 cases. In fact, in some cases there was a

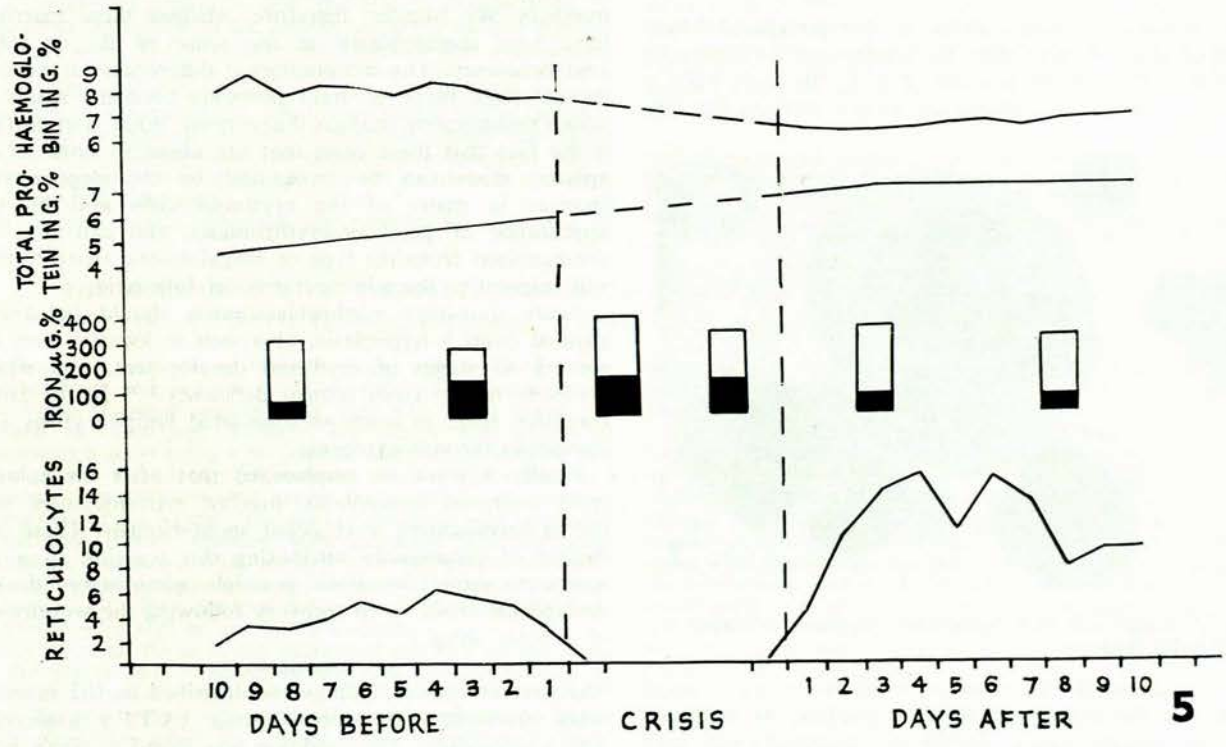

Fig. 5. Group B. Average values of 17 cases of acute transitory erythroblastopenia, including the serum total protein.

thrombocytosis and this finding, in association with an absence of the polychromatic cells, occasionally led to the diagnosis.

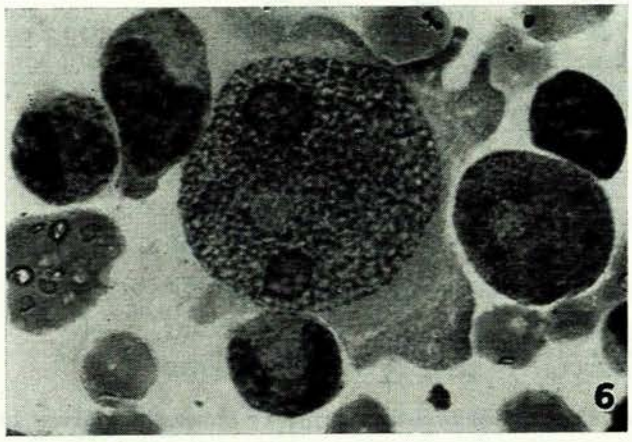

Fig. 6. Giant reticulum cell (cell 42·5μ), nucleus (26·5μ).

Recovery Phase

Marrow aspirations were studied from 40 cases during the recovery phase. The most obvious sign of recovery (which occurred without specific therapy) was the reappearance of numerous proerythroblasts. After this, all members of the erythroid series appeared in sequence, and an erythroid hyperplasia occurred. In all but one of the cases studied, maturation was normoblastic in type. It is of interest that some of these cases had been regarded as megaloblastic during the degenerative phase (Figs. 2A and 2D).

An increase in reticulocytes in the peripheral blood followed about 4 days after the appearance of numerous proerythroblasts in the marrow (Fig. 5). In some cases a granular leukocytosis, sometimes with a shift to the left,

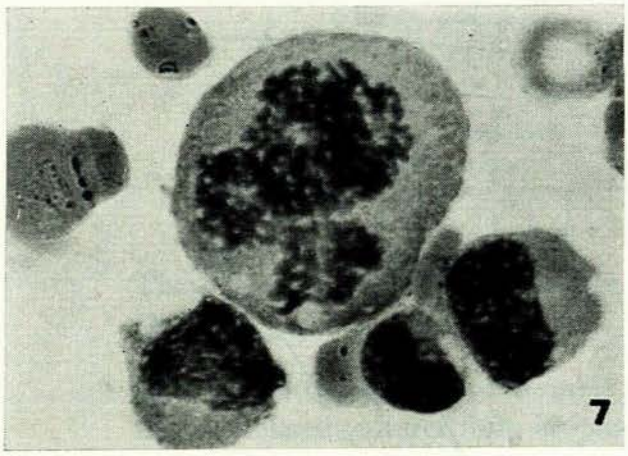

Fig. 7. Large cell with basophilic cytoplasm, showing a multipolar mitosis.

and an increase in the number of platelets, were noted just before the increase in the reticulocytes. At the same time, in certain cases, numerous nucleated red cells appeared in the blood; in one case the total number of nucleated cells was as high as 60,000 per cu.mm., of which 23,000 per cu.mm. were nucleated red cells. The presence of nucleated red cells and of the early granular series was of variable but usually short duration, and smears taken daily showed marked differences both in type and number of cells. The features were compatible with those that follow increased marrow stimulation.

2. **Serum Iron and Marrow Haemosiderin**

Serum-iron values were usually high during the erythroblastopenic phase, but dropped, often to very low levels, during recovery from the aplastic crisis (Fig. 5). The marrow haemosiderin gradings corresponded.

COMMENT

Patients about to undergo aplastic crises can be recognized by the appearance in the bone marrow of giant proerythroblasts, and by the degenerative changes in many of the other erythroid cells, whether normoblastic or megaloblastic in type. It is probable that the giant proerythroblast is only present during an aplastic crisis; we have never noted this type of cell in any other condition. It is probably a polyploid cell[14] that has undergone endomitosis and, thus failing to divide, has doubled in size.

It is of interest that some of the cases that have been regarded as megaloblastic during the degenerative phase have shown a normoblastic hyperplasia during the recovery phase of the aplastic crisis. The nucleus of this type of megaloblast has usually shown irregular clumping of the chromatin and has differed from the scroll-like pattern of the typical megaloblast. In addition, changes in the granular series have not been marked, and in most cases the platelets have been present in normal or increased numbers. We wonder, therefore, whether these marrows have been megaloblastic in the sense of B_{12} or folic-acid deficiency. The morphological differences in the erythroid series, however, have probably been the result of added degenerative changes (karyolysis). What is important is the fact that these cases that are about to undergo an aplastic crisis can be recognized by the degenerative changes in many of the erythroid cells and by the appearance of giant proerythroblasts, and can thus be distinguished from the type of megaloblastic marrow that will respond to the administration of folic acid.

Acute transitory erythroblastopenia should be distinguished from a hypoplasia, also seen in kwashiorkor, involving all stages of erythroid development, and which probably results from protein deficiency.[2, 15] In the latter condition there is often an associated lymphocytosis and sometimes thrombocytopenia.

Finally it must be emphasized that after an aplastic crisis erythroid hyperplasia, marked reticulocytosis, and rise in haemoglobin level, occur spontaneously. There is a danger of erroneously attributing this reaction to a response to some haematinic principle administered during the aplastic crisis, or to recovery following the withdrawal of a 'toxic' drug.

SUMMARY

The haematological findings are described in 102 cases of acute transitory erythroblastopenia (A.T.E.) associated with kwashiorkor. The condition was found to occur both in normoblastic and megaloblastic marrows, and consisted

of a degenerative phase, an aplastic phase, and a recovery phase. During the degenerative and aplastic phases giant proerythroblasts (25 - 50μ) and giant reticulum cells (40 - 60μ) were usually present and, apart from an occasional proerythroblast, members of the erythroid series disappeared from the bone marrow. The peripheral blood was characterized by a reticulocytopenia of about 10 days' duration. It was possible to differentiate the megaloblastic marrows that were about to undergo an aplastic crisis from those that would respond to folic-acid therapy.

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