ABNORMAL ERYTHROPOIESIS AND EOSINOPHILIA
REPORT OF TWO CASES OF EOSINOPHILIA ASSOCIATED WITH EITHER ERYTHROBLASTOSIS FETALIS OR INFANTILE HYPOPLASTIC ANEMIA

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Eosinophilia is a rather common condition observed in various diseases. Among the diseases of the hemopoietic system, this condition is well noted in chronic myelocytic leukemia, erythremia, Hodgkin's disease, eosinophilic leukemia, pernicious anemia, sickle cell anemia, and post-splenectomy(1). I have observed two cases of eosinophilia associated with either erythroblastosis fetalis or infantile hypoplastic anemia, which are of great interest and deserve detailed description with special reference to the relationships between abnormal erythropoiesis and eosinophilia.

REPORT OF CASES

Case 1
Clinical picture. The subject was a boy, who was born in the thirty-seventh week of gestation. His mother had given birth to two healthy boys, and her blood picture was normal (eosinophils 1 per cent). The new-born child showed pronounced abdominal distention. The body-weight at birth was 3,470 gm. The placenta was edematous, weighing 660 gm. After birth he cried feebly two times, henceforward fell into asphyxia and died thirty minutes after delivery. Both he and his parents showed negative Wassermann reaction. The results of blood typing were as follows: Father, Rh₉ positive, O, N; mother, Rh₃ positive, B, M; infant, Rh₀ positive, B, MN.

Hematologic findings. The blood from umbilical cord and the cardiac blood were examined. The red cell count was from 2.67 to 3.16 million per c. mm., the hemoglobin 78 per cent (Sahli), and the leukocyte count from 8,500 to 13,900 per c. mm. The outstanding features in the differential count of all the nucleated cells were the marked increase in the erythroblasts (29.5 to 33.5 per cent) and the eosinophils (30.5 to 34.0 per cent). The majority of the erythroblasts were polychromatic or basophilic, and a few erythroblasts in mitosis were found, but no megaloblast was noticed. More than half of the eosinophils were segmented (18.5 to 22.0 per cent), and it was not seldom to find eosinophils with three or four lobes. The immature eosinophils (myelocytes and promyelocytes) were also found in 4.5 to 8.5 per cent. Their oxydase reaction as well as the peroxydase reaction were intensively positive. The myeloblasts were present in 1.5 per cent.

The smears, which were made from the liver, spleen and the bone marrow of femur at the post-mortem examination, were stained with May-Grünwald-Giemsa stain, and the
nucleated cells, excluding the liver cells, reticular cells and endothelial cells, were examined in differential count. In either organs, both the erythroblasts and the eosinophils were markedly increased (Fig. 1) as follows: in the liver, the former being 52.0 per cent and the latter being 36.5; in the spleen, 29.5 and 46.0; in the bone marrow, 35.5 and 62.0. The basophilic and polychromatm erythroblasts (normoblasts) were far more numerous than the orthochromatm. Many of the eosinophils, comparing with that of the peripheral blood, were in immature type (myelocytes and promyelocytes). The myelogram, according to Rohr, of the femur also indicated the great increase of both the eosinophils (93.5 per cent) and the erythroblasts (43.5 per cent). The increase in number of the eosinophilic myelocytes (41 per cent) in the former and of the polychromatm and basophilic erythroblasts (28 per cent) in the latter was the most outstanding manifestation.

Pathologic findings (Autopsy No. 3211).

The abdomen was markedly distended and the face cyanotic. Marked and extensive hydrops and pink-yellowish effusion (300 cc.) in the peritoneal cavity, and a few scattered petechiae in the serosa and mucosa were noticed. The cardiac blood was unclotted. No signs of jaundice was observed.

The liver was considerably enlarged and weighed 170 gm. Microscopic examination revealed numerous eosinophils, mostly being eosinophilic myelocytes, and erythroblasts both of which were scattered diffusely or in islands in the lobules (Fig. 2). Consequently the sinusoids were dilated, and some liver cells were compressed. In the liver cells only a very few bile thrombi, fatty droplets, and hemosiderin-deposits could be seen. A fairly diffuse increase of the reticular fibers was noticed in the lobules, but either no spirochetes, granuloma, necrosis, or hemorrhages were demonstrated. In the portal spaces almost no sign of cell infiltration was seen and the bile ducts and vessels showed no abnormalities.

The spleen was greatly enlarged, weighing 26 gm. The reticulum of the pulp cords was moderately expanded, and packed with great numbers of eosinophils (mostly eosinophilic myelocytes) and erythroblasts. Neither Charcot-Leyden crystals nor fibrosis could be seen. The malpighian bodies were very small. In accordance with the eosinophilia in the red pulp, there were eosinophils in the subendothelial spaces of the trabecular venules (Fig. 3). The deposits of hemosiderin could be scarcely noticed.

The bone marrow of the femur was red and hyperplastic. Bone marrows of rib, vertebra and femur revealed the same microscopic pictures as follows: eosinophilic myelocytes and erythroblasts were noticed predominantly and diffusely increased, replacing the fatty tissues of the marrow (Fig. 4), while the mega-karyocytes decreased in number considerably. There was no vascular changes, hemosiderin, or Charcot-Leyden crystals. The osseous tissues showed no gross
ABNORMAL ERYTHROPOIESIS AND EOSINOPHILIA

changes, nor osteochondritis syphilitica.

The granules of all these eosinophils were stained intensively by the Nadi-solution or the 40 per cent alcohol-Sudan III colloidal solution (Kawamura-Yazaki’s stain(2)) (Fig. 3).

The heart was enlarged, weighing 31.5 gm. The foramen ovale was not closed. The subepicardial tissue was somewhat infiltrated with eosinophilic myelocytes and erythroblasts.

The lymph nodes showed no enlargement and no hematopoiesis.

The adrenals weighed left 4.3 gm. and right 3.4 gm. From medulla to zona fasciculata marked hyperemia which was accompanied by the marked hyperplasia of erythroblasts was noticed, but the eosinophils were not abundant. The degenerative changes in the cortex were not seen.

Case 2

Clinical picture. The patient was a female infant, born June 10, 1950, a month before term. Her birth weight was 2,250 gm. The family history was no contributory; her sister and brother showed no abnormalities hematologically. The infant had an adequate diet and developed normally till the end of August, at that time the temperature increased to 39°C continuing one week. She was given orally 1.5 gm. of sulfadiazine daily for these seven days. Thereafter she passed green watery stools for about two weeks. About the end of September anemia was noticed and this condition increased gradually. From Oct. 17 the temperature again rose up to 38° or 39°C and anemia increased moreover, and on Oct. 30 the patient expired. There was neither hemorrhagic diathesis, nor enlargement of the liver, spleen and lymph nodes. Urinalysis was negative; in the stool there was no egg of parasites. The patient and the parents showed negative Wassermann reaction.

Hematologic findings. The blood pictures on Oct. 3, 12, 18, 23 and 28 were as follows: The red cell count and the hemoglobin decreased markedly in the course of the disease, the former being 4.1, 2.4, 2.15, 2.2 and 1.2 million per c. mm., the latter 68, 48, 23, 17 and 14 per cent (Sahli), respectively. The leukocyte count, however, rose gradually from 9,000 to 9,900, 17,000, 27,000 and 22,000. The outstanding feature in the differential count of the nucleated cells was the conspicuous increase in the eosinophils, which percentages were 35.0, 45.5, 63.0, 61.0 and 29.0 per cent, respectively. The immature red cells were only rarely seen: that is, the normoblasts count was always limited between 1.0 and 1.5 per cent and the polychromatric erythrocytes were only seldom seen in each above-mentioned blood pictures. Myeloblasts and myelocytes were seen only a few at times (0.5 per cent). Now, more than half of the markedly increased eosinophils had two nuclei, but it was not seldom to find eosinophils with three or four lobes. The eosinophilic myelocytes and metamyelocytes, however, were scarcely noticed. The cytoplasm and its granules appeared normal. The myelogram of the sternum on Oct. 17 disclosed the marked decrease of erythroblasts (7 per cent), especially orthochromatic erythroblasts (1 per cent). Megaloblasts were not seen. Many of the eosinophils, which were markedly increased to 77 per cent, had the nuclei of two lobes (40.5 per cent), and the eosinophilic myelocytes and metamyelocytes, however, were also found in great numbers (19.5 per cent) (Fig. 5).
Pathologic findings (Autopsy No. S-29).

The body and the various viscera of this 5-month-old infant was markedly anemic, without hydrops, petechiae, jaundice, and rash.

The liver was not enlarged, weighing 155 gm. Microscopically, the sinusoids were slightly dilated with the plasma-like materials and a very few red cells. In the sinusoids, there were no hematopoietic foci, except for a few eosinophils being scattered. Some Kupffer cells contained Charcot-Leyden crystals. The central fatty degeneration of acini caused by anemia was conspicuous. In the portal spaces the mononuclear or polymorphonuclear eosinophils infiltration was strong. Inflammatory changes, hemorrhages, hemosiderin and fibrosis were not noticed.

The spleen was not enlarged, weighing 15 gm. The follicles were somewhat atrophic and many of them had Flemming's centers. In the central fields of the Flemming's centers the network, which is composed of swollen reticulum cells and their fibrillar parts, was expanded and packed loosely with a few lymphocytes and epithelioid cells, both of which nuclei showed karyorrhexis markedly ("Reticular form" of Ono and Nakagawa(3)). In the pulp cords the infiltration of eosinophils was conspicuous and the eosinophilic myelocytes were numerous. Some of the reticulum cells contained Charcot-Leyden crystals. Erythroblasts and hemosiderin-deposits were not seen.

The bone marrow of the femur was grey-red and hyperplastic throughout. Microscopically, the findings of that of rib, vertebra, and femur were all the same: These were cellular and hyperplastic. The cellular increase was found to be due to the pronounced proliferation of eosinophils (Fig. 6), and their myelocytes were also noticed in great numbers. In contrast to the eosinophilia, the erythroblasts were sharply reduced, and took the loosely scattered position without forming cellular islands. On the other hand, the megakaryocytes showed no significant changes. There were many swollen reticulum cells containing Charcot-Leyden crystals. No hemosiderin, fibrosis, nor syphilitic osteochondritis was noticed.

The lymph nodes showed no enlargement. The majority of the follicles presented the similar changes to that of the spleen. The pulp cords were infiltrated with eosinophils considerably.

The heart, which weighed 31.5 gm., was slightly enlarged without anomalies and presented slight edema around the small vessels in the myocardium. The kidneys disclosed cloudy swelling of the epithelium in the convoluted tubules. In the gastrointestinal tract there was no parasites, ulcer, or hemorrhages. In the endportion of the ileum, the follicles were also in the reticular form, and in
the submucosa a few eosinophils were scattered loosely. In the brain the perivascular spaces were slightly edematous. The lungs presented slight catarrhal bronchitis, but no signs of pneumonia or eosinophilia was seen.

Adrenals: The left weighed 2.3 gm., the right weighed 2.0 gm., and were normal in size. The lipid-contents and the appearance of the cortex cells were normal. No eosinophils infiltration was noticed. In the pituitary, which weighed 0.1 gm., the eosinophilic-type cells were swollen and increased slightly.

COMMENT

Case 1 was a new-born infant, which died thirty minutes after delivery, with the outstanding manifestations of hydrops, erythro-(leuko)blastosis, hepato-, and splenomegaly. Congenital syphilis or infantile erythroblastosis (Jaksch-Hayem's anemia and Cooley's anemia) etc., which is the cause of an increase in numbers of nucleated red cells, however, could be denied. Therefore this case should be regarded as erythro-(leuko) blastosis fetalis. The intralobular fibrosis, which was noticed in case 1, was also described by Henderson(4) and Potter(5) in the same disease.

In case 2, severe and progressive anemia, few immature erythrocytes in peripheral blood and the marked decrease in numbers of erythroblasts in bone marrow were the striking changes, and these pathologic conditions were not due to syphilis or parasitic infections. Consequently this case should be considered as a hypoplastic anemia or a primary refractory anemia as described by Wintrobe(1) and others.

The characteristic features in both cases 1 and 2 were the marked eosinophilia. In erythroblastosis fetalis, generally, the leukocytic series are also more or less hyperplastic(1,6,7). Especially in the cases of erythro-leukoblastosis fetalis described by Gierke(7), Salmonsein(8) and Wanstrom(9), leukocytes share more or less equally with the red cells in the medullary and extramedullary hematopoiesis. In formerly described cases of erythro-leukoblastosis and erythroblastosis fetalis, some eosinophils are occasionally found associated with other granulocytes in the blood and various organs, but are not predominant. Comparing this point with my case, I noticed that the eosinophils in case 1 were far more increased than in the other above-mentioned cases.

In case 2 familial eosinophilia can be denied by the family blood pictures. And this case can not be considered histologically as eosinophilic leukemia, which is characterized by the uncontrolled, destructive growth of the eosinophils.
In the fetal and infantile diseases of the hematopoietic system, the erythrocytic series and the leukocytic series, as a rule, are easily influenced by each other. It has been emphasized by some authors\(^7,^10\) that this point is characteristic in the fetal and infantile diseases of the hematopoietic system. By what mechanism is the implication in the both series caused? The relation between both series in these cases is too intimate to be considered as myeloid metaplasia in adult anemia or abnormal anemia in adult leukemia. In the cases of erythroblastosis fetalis, jaundice, Kernicterus, bile thrombi and hemosiderin-deposits have been noticed and considered as the result of disturbances in hemoglobin metabolism in wide sense. Furthermore, the staining in Kernicterus has been thought to be brought about by a pigment other than bilirubin\(^11\). Moreover such a abnormal substance as xanthorubin has been detected by Gierke\(^7\). From this point of view couldn’t my cases of eosinophilia be discussed?

First I want to discuss the relation between erythrocytes and eosinophils. Once Klein\(^12\) and Weidenreich\(^13\) surmised that eosinophile granules are composed of hemoglobin itself. This theory of the hemoglobinous origin of the granules, however, can not be approved nowadays experimentally\(^14\) and histochemically. In the diseases of erythrocytic system, the eosinophilia has been occasionally noted in the cases of erythremia (polycythemia vera\(^1,^15\)), pernicious anemia\(^1\) and hypoplastic anemia caused by bezene\(^16\). According to Ringoen\(^14\), Pescatori\(^17\) and others, eosinophilia is caused by anaphylaxis, allergy, or asphyxia, but the mechanism remains obscure. Döhnert and Tischendorf\(^18\) have pointed out that eosinophils are observed in large numbers around the erythroblastic foci in the widely extended red femur marrow, which occurs frequently in cases of liver cirrhosis, and suggested that eosinophile granules have intimate relations with hemoglobin or its precursors. Amano\(^19\) has recognized some relationship between eosinophile granules and hemoglobin ontogenetically and phyrogenetically, by arguments as follows: 1) In the 1.3 cm embryo, which is the beginning of the hepatic period of hematopoiesis, the eosinophile myelocytes can be demonstrated as the earliest of all the granulocytes in the liver, which are seen after the appearance of the erythroblasts of 2. generation\(^20\). 2) In some invertebrates, that have erythrocytes or hemoglobin definitely, are in possession of either eosinophile granulocytes or the peroxydase reaction positive granulocytes.

Next the relation of hemoglobin in erythrocytes, cytochrome, which is contained greatly within eosinophile granules according to Amano\(^21\), and peroxydase in eosinophiles, should be considered. According to Fischer and Zeile, either
Case 1. Numerous eosinophils and erythroblasts in the bone marrow smear of the femur.
May-Grünwald-Giemsa stain.

Case 1. Numerous islands of eosinophils (A) and erythroblasts (B) in the hepatic lobules.
Hematoxylin and eosin

Case 1. Numerous eosinophils in the splenic pulp. A very small malpighian body (A).
Eosinophils in the subendothelial space of the trabecular venule (B).
Kawamura-Yazaki’s stain.

OONEDA I
Fig. 4.
Case 1. Marked hyperplasia of erythroblasts and eosinophils in the bone marrow of the rib.
Hematoxylin and eosin stain.

Fig. 5.
Case 2. Numerous eosinophils in the sternal marrow smear.
May-Grünwald-Giemsa stain.

Fig. 6
Case 2. Pronounced proliferation of eosinophils and sharply reduced and loosely scattered erythroblasts in the bone marrow of the femur.
Hematoxylin and eosin stain.
hemoglobin, cytochrome, or peroxidase has a porphyrin as a prosthetic group, and the porphyrin found in the hemoglobin and cytochrome is classified as etioporphyrin type III similarly. Therefore the component part of cytochrome or peroxidase is common to that of hemoglobin. Accordingly, I suppose that in the hypoplastic anemia (a suppression of regeneration) and in the erythroblastosis fetalis (a suppression of maturation in spite of hyperplasia) the hemoglobin-synthesis is disturbed and consequently porphyrin is accumulated in the bone marrow and the increased porphyrin, on the other hand, is utilized excessively to the formation and maturation of eosinophile granules, leading up to the development of the eosinophilia.

SUMMARY

Two cases of marked eosinophilia accompanied by either erythro-(leuko)-blastosis fetalis or infantile hypoplastic anemia have been reported with hematologic and pathologic descriptions.

The former case was characterized by the marked erythroblastosis in various viscera, and the latter case by the marked suppression of erythropoiesis, both cases presenting alike marked eosinophilia in the bone marrow, spleen, liver and blood.

The intimate relationships between the abnormal erythropoiesis and eosinophilia in these cases are considered as the results of the disturbed metabolism of porphyrin, which is the common component of the cytochrome in the eosinophile granules and the hemoglobin in erythrocytes.

REFERENCES