Effect of Age on Hematopoiesis in Man

By D. A. Lipschitz, K. B. Udupa, K. Y. Milton, and C. O. Thompson

We have shown previously that the cause of anemia in healthy elderly subjects can usually not be identified. In this study, hematopoiesis was examined in 18 healthy elderly subjects with unexplained anemia and in 15 young and 15 healthy elderly individuals without anemia. No reduction in circulating testosterone was noted, making decreased androgen levels as a cause for the anemia unlikely. The 2,3 diphospho-glycerate (2,3DPG) levels in the anemic subjects were significantly higher than their corresponding controls, suggesting that the anemia was pathologic, as no increase would be expected if the low hemoglobin was a physiologic adjustment to age. The anemia was associated with a reduction in marrow normoblast and CFU-E number, but no decrease in BFU-E levels was seen. This suggests that the mechanism of the anemia is a decrease in stem cell proliferation. This could be caused by a reduction in circulating erythropoietin or a defect in end organ response. A second possibility is that a basic cellular abnormality exists. The presence of an overall reduction in hematopoiesis in anemic elderly (decreased peripheral blood counts, reduced marrow myeloid precursors, and CFU-C levels) makes this especially likely. The abnormality may be caused by a mechanism unrelated to the aging process. The fact that nonanemic elderly also have reductions in hematopoiesis suggests that age contributes to the defect.

LITTLE INFORMATION is available regarding the effects of aging on bone marrow function. One area that has been extensively investigated has been anemia. Large epidemiologic studies in the United States and Canada indicate that anemia in females above age 59 is approximately equal to that found in females of child-bearing age. In men, a definite increase in the older age group is found. Studies from Great Britain are important, as they have determined the incidence of anemia in large numbers of subjects over age 60. In both males and females, the incidence of anemia increases significantly with each successive decade. Anemia is not thought to be a normal feature of aging, as the etiologies in the elderly are very similar to those in young subjects. Thus, iron deficiency and the anemia of chronic disease are quoted as the two most frequent diagnoses. Recently, however, a careful evaluation of healthy elderly living at home showed that iron deficiency anemia is much rarer in elderly than in young females and accounts for only a small fraction of anemia in elderly males. We have recently demonstrated a high incidence of anemia in a group of carefully selected healthy elderly subjects living in the Little Rock, AR, area. We confirmed that iron deficiency anemia was rare and found that the anemia of chronic disease, folate deficiency, hemolyis, and other rarer causes of anemia were uncommon. The elderly males and females with unexplained anemia had significantly lower leukocyte and neutrophil counts than nonanemic subjects, suggesting mild marrow failure. The present study was undertaken to examine in greater detail the effect of age on hematopoiesis in man. Groups of young and elderly nonanemic subjects were studied, and their results were compared to the findings in healthy elderly with unexplained anemia. We measured testosterone, 2,3 diphospho-glycerate (2,3DPG) levels, and quantitated hematopoietic precursors by ferrokinetic measurements and histologic examination of bone marrow biopsies.

MATERIALS AND METHODS

Studies were performed on 30 hematologically normal white males, 15 under the age of 45 (mean 34 yr) and the remainder over the age of 70 (mean 78 yr). Eighteen white male subjects above the age of 70 (mean age 82 yr) with a mild unexplained anemia (hemoglobin less than 13 g/dl and hematocrit of less than 39%) were also examined. In all subjects, multiple measurements were performed and the hemoglobin and hematocrit remained stable. All the young healthy subjects were volunteers recruited at the University of Arkansas for Medical Sciences or subjects admitted to the Veterans Administration Hospital for minor elective surgery. Five of the nonanemic and three of the anemic elderly were also admitted for elective surgery. The remaining subjects were volunteers surveyed for anemia among elderly attending a variety of community activities in the greater Little Rock area. None gave a history of an acute illness within the last 3 mo, and they were all a pyrexial at the time of study. A careful history and physical examination was performed on each. None had a disease known to affect hematopoiesis (cancer, chronic infections, collagen vascular disease, rheumatoid disease, etc.), and physical examination failed to reveal any abnormalities associated with abnormal hematopoiesis (splenomegaly, purpura, clubbing, etc.). Subjects with diabetes were excluded, as were any individuals taking prescription drugs (including antihypertensives and diuretics) for any reason. Many of the subjects, both young and old, took occasional “over the counter” medications, including analgesics and laxatives. Five of the young and seven of the elderly subjects took a multivitamin preparation daily. Myxedema was excluded by the measurement of T4, T3, and thyroid-binding globulin. Evidence of liver disease and/or renal disease, either by history or by routine laboratory procedure, also made the subject...
in eligible for study. Since protein calorie malnutrition affects hematopoietic function, subjects were excluded if their serum albumin was less than 3.5 g/dl.

The criteria for defining a subject as having unexplained anemia included: (A) No evidence of iron-deficient erythropoiesis, as determined by a mean cellular volume (MCV) > 84 fl, serum iron > 60 μg/dl, transferrin saturation > 20%, and a red blood cell protoporphyrin/heme ratio < 30 μmol/mole. (B) Normal iron stores as determined by serum ferritin > 20 ng/ml, adequate stores on histologic examination of bone marrow, and normal sideroblast number. The total iron binding capacity (TIBC) was < 400 μg/dl but > 250 μg/dl (a level below 250 μg/dl would suggest malnutrition or chronic disease and a value greater than 400 μg/dl would suggest deficient iron stores). (C) Normal serum B12 and serum and red cell folate levels. (D) A reticulocyte production index less than 2.5 and an indirect bilirubin less than 0.6 mg/dl. (E) A normal peripheral smear examination. (F) Morphological examination of bone marrow aspirate and biopsy revealed no cause for the anemia. To obtain the 18 anemic subjects studied, a total of 195 healthy elderly males were surveyed. Of these, 38 (19.5%) had a hemoglobin < 13 g/dl and hematocrit < 39%, and 26 (66%) of the anemic group met the rigid criteria enumerated above. Of these, 18 voluntarily agreed to participate in this study, which was approved by the Human Research Committee of the University of Arkansas for Medical Sciences and the Little Rock VA Hospital.

In each of the subjects examined, marrow normoblast number was quantitated by ferrokinetic measurements, as described in detail by Finch et al. 10 Marrow normoblasts number (normal value 5.4 × 10^9/kg) is calculated from the erythron iron turnover, which is derived from the plasma iron turnover. The details of measurement of the plasma iron turnover (PIT), erythron iron turnover (EIT), and the calculation of marrow normoblast numbers have been extensively reported elsewhere.10–12 The validity of this method of quantitating erythroid precursors has been established by the study of both normal subjects and patients with a wide range of disorders of erythropoiesis.12–13 It must be emphasized that marrow normoblast number is determined by ferrokinetics, which estimates erythroid mass, and the results are expressed/kg body weight for convenience only. The actual weight of the subject studied is not used in the calculation.

Marrow Culture

Two to three milliliters of marrow was aspirated from the iliac crest and transferred to a sterile polystyrene tube containing 0.1 ml preservative-free sterile heparin. After appropriate washing, the sample was diluted so that 5 plates, each containing 10^6 nucleated cells, were cultured in vitro to quantitate the number of committed macrophage/granulocyte progenitor cells (CFU-C). The method of Pike and Robinson14 was used, except that a feeding layer was not employed. Instead, peripheral white cell conditioned medium was used as a source of colony-stimulating activity.15 Colonies, defined as aggregates of greater than 40 cells, were scored at day 14. In some studies, 15 plates, containing 2 × 10^5 nucleated cells, were cultured in methylcellulose for erythroid burst-forming units (BFU-E) and for the erythroid colony-forming unit (CFU-E) using the method described by Gregory and Eaves.16 CFU-E were scored 8 or 9 days after plating. Using the criteria of Gregory and Eaves, small bursts (mature BFU-E) were scored on day 12 and large bursts (immature BFU-E) were scored on day 20. For CFU-E culture, 0.4 U Step III erythropoietin (Ep), purchased from Connaught Laboratories, was added to each culture and 2 U Ep was added to each for the BFU-E assays.

In order to perform differential counts, multiple aliquots of the marrow suspension used for plating were cytocentrifuged and the cytoospin preparations stained with Wright’s stain. The number of progenitor cells (CFU-C, BFU-E, CFU-E) was then quantitated, assuming that each colony was derived from a single committed progenitor cell. Once the colonies had been scored, the following formula was used to calculate the progenitor cells/kg, as described previously:17

\[
S_T = \frac{S}{N} \times \frac{E}{(1 - R)}
\]

where \(S_T\) = number of progenitor cells/kg body weight; \(S\) = number of colonies cultured; \(N\) = number of nucleated cells plated; \(E\) = number of marrow normoblasts × 10^9/kg body weight; and \(R\) = ratio of normoblasts to total nucleated cells in marrow. This was determined by differential counts of 10 × 10^3 cells on cytoospin preparations of the sample plated.

In the studies described in this article, marrow aspirated from the iliac crest was used to culture committed progenitor cells. Because a redistribution of marrow has been reported with age,18 preliminary studies were undertaken to validate the use of iliac crest marrow in the elderly. This was done by the simultaneous culture and differential counts of marrow aspirated from the iliac crest and sternum of 6 volunteers above age 65. The mean ± SE CFU-C/10^6 cells was 46 ± 4 and 44 ± 6 for the iliac crest and sternum cultures, respectively. In 3 subjects, CFU-E was also measured, with the mean (range) being 41 (36–46) colonies/2 × 10^5 cells from the sternum and 44 (32–53) colonies/2 × 10^5 cells from the iliac crest. Differential counts of cytoospin preparations revealed a ratio of erythroid:nucleated cells in the sample plated of 0.16 ± 0.01 from the iliac crest and 0.17 ± 0.01 from the sternum. These results demonstrate that the proportion of hematopoietic cells aspirated from the sternum and iliac crest marrow in elderly subjects is identical and indicate that the use of iliac aspirates provides a valid reflection of progenitor cells measured.

Marrow Biopsy

Marrow biopsies were obtained from the posterior iliac crest with a Jamshidi needle and prepared for histology as follows. They were fixed in 5% Acrolein, decalcified, and embedded in methacrylate as described previously.19 Sections 2-μm thick were prepared using a JB microtome and were stained with 1% eosine-Y and 0.1% azure II after mounting and removal of the plastic. All sections were examined histologically, and the ratio of erythroid:myeloid precursors was determined by counting 10 × 10^6 cells in multiple sections cut at least 12 μm apart. This avoided the problem of multiple cell counting. Previously reported correction factors were used to adjust crude erythroid:myeloid ratios.13

CBC, differential count, platelet count, and reticulocyte count were measured using standard hematologic procedures. Serum iron and TIBC were measured using the methods recommended by the International Committee for Standardization in Hematology. Serum ferritin was determined by a "2-site" immunoradiometric assay, as described by Miles et al.22 The red blood cell protoporphyrin to heme ratio was determined using the method of Labbe et al.21 Serum B12 and folate were measured using the quanta-count kit method (Becton Dickinson, Rutherford, NJ). Red cell 2,3-DPG was determined using a commercial kit purchased from Sigma Laboratories (St. Louis, MO) and serum testosterone was measured by an assay developed by Diagnostic Products (Los Angeles, CA).

RESULTS

The hemoglobin values and hematologic profiles of the young and elderly controls and the elderly anemic controls were determined by the criteria noted above. The results are summarized in Table 1. The age distribution of the subjects is shown in Figure 1. As expected, the mean age of the young controls was 30 years (range, 20–35 years), and the mean age of the elderly controls was 80 years (range, 71–86 years). The mean (±SE) Hb level of the young controls was 13.6 ± 0.2 g/dl (range, 12.5–14.5 g/dl), and the mean (±SE) Hb level of the elderly controls was 10.2 ± 0.2 g/dl (range, 8.5–12.0 g/dl). The mean (±SE) hematocrit of the young controls was 40.2 ± 1.2% (range, 35.0–45.0%), and the mean (±SE) hematocrit of the elderly controls was 30.5 ± 1.2% (range, 24.0–36.0%). The mean (±SE) platelet count of the young controls was 285 ± 22 × 10^9/l (range, 200–350 × 10^9/l), and the mean (±SE) platelet count of the elderly controls was 270 ± 20 × 10^9/l (range, 200–300 × 10^9/l). The mean (±SE) reticulocyte count of the young controls was 1.1 ± 0.1 × 10^9/l (range, 0.8–1.5 × 10^9/l), and the mean (±SE) reticulocyte count of the elderly controls was 0.6 ± 0.1 × 10^9/l (range, 0.3–0.9 × 10^9/l).
In order to determine whether or not the unexplained anemia seen in the elderly subjects was merely a physiologic adjustment to decreased red cell requirements or rather represented a real abnormality, 2,3DPG levels were measured in 10 individuals from the elderly anemic group and in 7 each from the young and old control groups. The mean (±1 SEM) 2,3DPG level in the young controls was 14.6 ± 1.1 μmol/g hemoglobin and 13.8 ± 0.4 μmol/g hemoglobin in the elderly controls. In the elderly anemic group, the value was 15.0 ± 0.3 μmol/g hemoglobin, which is significantly higher than in the old controls (p < 0.01).

The mean (±SE) marrow normoblast number, determined by ferrokinetic measurements, of 3.14 ± 0.24 x 10⁶ cells/kg in the elderly anemic subjects was significantly less (p < 0.001) than a mean of 4.82 ± 0.26 x 10⁶/kg in the young controls and was also lower (p < 0.03) than a mean of 4.32 ± 0.36 x 10⁶/Kg in the elderly without anemia. Erythroid progenitor cells were quantitated in 7 young and 5 old controls and in 6 anemic elderly. The number of CFU-E/2 x 10⁵ cells averaged 41 colonies in the elderly anemic group, which was significantly lower than means of 100 colonies/2 x 10⁵ cells in the young controls (p < 0.001) and 68 colonies/2 x 10⁵ cells in the old controls (Table 2). From these values, marrow normoblast number and the ratio of erythroid to total nucleated cells in the sample plated CFU-E/kg was calculated. The mean number in the elderly anemic group of 0.36 x 10⁶ cells/kg was significantly less (p < 0.001) than values of 1.08 x 10⁶ cells/kg in the young controls and 0.75 x 10⁶ cells/kg in the elderly controls: 34.0 (2.0) mg/dl in the young controls; and 26 ng/dl in the elderly controls and was also lower (p < 0.03) than a mean of 4.32 ± 0.36 x 10⁶/Kg in the elderly without anemia.

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### Table 1. Hematologic Profile in Young and Elderly Subjects With Unexplained Anemia

<table>
<thead>
<tr>
<th>Age (yr)</th>
<th>Young Control</th>
<th>Elderly Anemic</th>
<th>Control</th>
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<tbody>
<tr>
<td>34.0 (2.0)*</td>
<td>82.0 (2.0)</td>
<td>78.0 (2.0)</td>
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<tr>
<td>15.4 (0.3)</td>
<td>11.5 (0.3)</td>
<td>15.0 (0.2)</td>
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<tr>
<td>89.0 (0.9)</td>
<td>94.4 (1.3)</td>
<td>90.7 (1.8)</td>
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<tr>
<td>107.0 (8.0)</td>
<td>91.0 (4.0)</td>
<td>93.0 (5.0)</td>
<td></td>
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<tr>
<td>297.0 (10.0)</td>
<td>289.0 (7.0)</td>
<td>307.0 (13.0)</td>
<td></td>
</tr>
<tr>
<td>36.0 (3.0)</td>
<td>31.6 (1.8)</td>
<td>30.1 (2.2)</td>
<td></td>
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<tr>
<td>126.0 (17.0)</td>
<td>234.0 (24.0)</td>
<td>219.0 (26.0)</td>
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<tr>
<td>24.4 (1.4)</td>
<td>22.1 (1.4)</td>
<td>22.0 (1.8)</td>
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<tr>
<td>476.0 (34.0)</td>
<td>521.0 (79.0)</td>
<td>461.0 (34.0)</td>
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<tr>
<td>5.6 (0.8)</td>
<td>6.6 (0.7)</td>
<td>4.8 (0.5)</td>
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<tr>
<td>1.1 (0.3)</td>
<td>0.8 (0.2)</td>
<td>1.0 (0.2)</td>
<td></td>
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<tr>
<td>8.8 (0.4)</td>
<td>5.1 (0.3)</td>
<td>7.6 (0.5)</td>
<td></td>
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<tr>
<td>5.9 (0.3)</td>
<td>3.4 (0.4)</td>
<td>4.6 (0.6)</td>
<td></td>
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<tr>
<td>1.9 (0.8)</td>
<td>1.1 (0.1)</td>
<td>1.9 (0.3)</td>
<td></td>
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<tr>
<td>361.0 (38.0)</td>
<td>227.0 (18.1)</td>
<td>277.0 (21.1)</td>
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</table>

*Mean (± 1 SEM) is shown.
†Elderly anemic and elderly controls significantly greater than young control (p < 0.001).
‡Elderly anemic significantly less than young control (p < 0.01).
§Elderly anemic significantly less than young control (p < 0.03).
‖Elderly anemic significantly less than young control (p < 0.004).
¶Elderly anemic significantly less than young control (p < 0.02).
Table 2. Erythroid Progenitor Cells (BFU-E, CFU-E) Expressed per 2 × 10⁶ Cells Plated and per Kilogram Body Weight in Young and Elderly Control Subjects and Elderly With Unexplained Anemia

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</thead>
<tbody>
<tr>
<td>Elderly anemic</td>
<td>45 67 33</td>
<td>2.91 0.12</td>
<td>0.46 0.34</td>
<td>0.15 0.38</td>
<td>0.38 0.50</td>
<td>0.02 0.34</td>
<td>0.45 0.38</td>
<td>0.02 0.38</td>
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<td>0.45 0.38</td>
<td>0.02 0.38</td>
<td></td>
</tr>
<tr>
<td>Elderly controls</td>
<td>50 61 87</td>
<td>4.69 0.16</td>
<td>0.61 0.74</td>
<td>0.11 0.63</td>
<td>0.81 0.44</td>
<td>0.02 0.34</td>
<td>0.55 0.74</td>
<td>0.04 0.38</td>
<td>0.55 0.74</td>
<td>0.04 0.38</td>
<td>0.55 0.74</td>
<td>0.04 0.38</td>
<td></td>
</tr>
<tr>
<td>Young controls</td>
<td>48 43 84</td>
<td>4.16 0.17</td>
<td>0.47 0.42</td>
<td>0.13 0.59</td>
<td>0.59 0.72</td>
<td>0.03 0.34</td>
<td>0.41 0.78</td>
<td>0.05 0.38</td>
<td>0.41 0.78</td>
<td>0.05 0.38</td>
<td>0.41 0.78</td>
<td>0.05 0.38</td>
<td></td>
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</tbody>
</table>

*Normoblast number × 10⁹/kg.
†R, ratio of erythroid:total nucleated cells in cytospin of sample plated.
‡Elderly anemic significantly less than young and elderly controls (p < 0.001).

2, Fig. 1). Although the mean value was lower in the old controls as compared to the young, the difference was not statistically significant whether or not the CFU-E was expressed per 2 × 10⁶ cells or per kilogram body weight. In contrast to CFU-E, which were lower in the elderly anemic group, both mature and immature BFU-E were not significantly different in the 3 groups studied (Table 2, Fig. 1), the mean for immature BFU-E/2 × 10⁶ cells being 45, 49, and 52 BFU-E in the young controls, elderly controls, and elderly anemic groups, respectively (Table 2). Corresponding values for mature BFU-E were 63, 58, and 67/2 × 10⁶ cells in the 3 groups, respectively. When expressed per kilogram body weight, both mature and immature BFU-E were not significantly different in the 3 groups studied (Table 2).
controls, but the differences were not statistically significant. The mean (±1 SE) number of promyelocytes/CFU-C averaged 79 ± 9 cells in the young control group, 77 ± 12 cells in the elderly controls, and 118 ± 18 cells in the elderly anemic group. All these values were not significantly different from each other. Similarly, the ratio of each myeloid precursor to the next more mature myeloid cell was the same in all groups studied. In the elderly anemic group, the more primitive precursors were the most suppressed, so that when total myeloid precursors/CFU-C was examined, the number was significantly higher in the elderly anemic group (p < 0.01) than young and old controls, the mean values being 1,624 ± 110, 913 ± 83, and 791 ± 103 total myeloid precursors/CFU-C, respectively.

DISCUSSION

The study of aged humans is made particularly difficult by the complex interrelationship between changes as a result of advancing age and those caused by dietary or environmental factors or the presence of overt or occult chronic diseases. This particularly applies to hematopoiesis, as many of these variables affect marrow function. For these reasons, very rigid criteria were used to select subjects for this study. Except for the presence of unexplained anemia in some, the elderly subjects were similar in all respects. By means of a careful history, physical examination, and a series of laboratory tests, these ambulatory, home-based elderly subjects could be defined as healthy. The young controls also met the rigid criteria required for the elderly group.

The prevalence of anemia in any group is based on hemoglobin and hematocrit measurement, which are the most convenient ways of detecting reductions in red cell mass. Because clear overlap occurs between subjects with low hemoglobin levels who are anemic and those who are not, the prevalence of anemia is usually expressed as the fraction of subjects at low risk, moderate risk, or high risk of being anemic. Virtually all the elderly subjects with unexplained anemia in this report had a mild abnormality, and hence, were at moderate risk of being anemic. Our studies indicate that the etiology of the anemia is not iron deficiency, chronic disease, or the other more commonly recognized causes. It is possible that these subjects have an appropriate hemoglobin and hematocrit value, as well as an appropriate red cell mass. Several mechanisms could account for a reduction in red cell mass with advancing age. In young subjects, red cell mass correlates very closely with lean body mass, the major determinant of oxygen requirement.24,25 A reduction in lean body mass occurs with age, and therefore, requirements for oxygen and red cells may decrease.26 Studies by Gardner and associates have shown that the correlation between red cell and lean body mass becomes less positive with age. This lack of correlation led these workers to postulate that reduced red cell mass in older subjects reflected a primary marrow abnormality.27,28

Testosterone has been shown to have a direct effect
on hematopoiesis and is generally accepted as the mechanism accounting for the difference in hemoglobin levels between males and females. In our study, no reduction in circulating hormone was found. However, an alteration in end organ receptor or response cannot be excluded. However, a recent study of healthy elderly has shown no reduction in testosterone, testosterone binding protein, or responsiveness of cell to the hormone with advancing age.29 The lower hemoglobin in elderly subjects may be caused by a decrease in circulating erythropoietin levels, but this has never been proven. A shortened red cell survival could also account for a reduction in red cell mass, but previous studies have shown normal survival in the elderly.30

In response to anemia, a predictable increase in 2,3DPG levels occurs. This increase in 2,3DPG averages 5% with each 1 g/dl drop in hemoglobin and results in a shift in the dissociation curve, allowing oxygen to be released more easily from hemoglobin to tissues.31 If the fall in hemoglobin seen in our elderly subjects with unexplained anemia reflects a normal response to age, tissue oxygen needs would not be increased and a rise in 2,3DPG levels would not occur. Our results confirm the previous finding that healthy nonanemic elderly have 2,3DPG levels that are modestly lower than young subjects.32 The elderly group with unexplained anemia had 2,3DPG levels significantly greater than the corresponding control group, strongly suggesting that the lowered hemoglobin is not a physiologic reduction due to the aging process. The 2,3DPG level in the elderly anemic group was, however, not significantly higher than the value noted in the young control group and was significantly lower than the levels usually found in younger subjects with a similar anemia from a variety of causes. This suggests that the 2,3DPG response to anemia may be inappropriately low in the aged or that oxygen requirements in the anemic elderly are lower than in young subjects with similar hemoglobin concentrations.

This study demonstrated that elderly individuals with unexplained anemia had significantly lower bone marrow erythroid precursors than the young or elderly controls. In addition, the erythropoietin-responsive erythroid progenitor cell, or CFU-E, was markedly reduced. In contrast, the more primitive erythroid progenitor cells (mature and immature BFU-E) were

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### Table 3. Hematopoietic Cell Numbers in Elderly With an Unexplained Anemia and Elderly Controls (the Mean ± 1 SEM for the Young Controls Is Also Shown)

<table>
<thead>
<tr>
<th>Total</th>
<th>Myelocytes</th>
<th>Metamyelocytes</th>
<th>Bands</th>
<th>Granulocytes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Erythron iron turnover</td>
<td>3.27±1</td>
<td>4.23±1</td>
<td>5.31±1</td>
<td>6.70±1</td>
</tr>
<tr>
<td>Mean</td>
<td>3.39±1</td>
<td>4.46±1</td>
<td>5.12±1</td>
<td>6.07±1</td>
</tr>
</tbody>
</table>

- Erythroid cell numbers are given as x 10^6 cells/kg.
- Myelocytes, promyelocytes, and myeloid CFU-C are given as x 10^6 cells/kg.
- Metamyelocytes, bands, and granulocytes are given as x 10^4 cells/kg.
- Total myeloid precursors are given as x 10^5 cells/kg.
- CFU-C are given as x 10^6 cells/kg.

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*Erythron iron turnover, mg/dl whole blood/day.

1 For each measurement shown elderly anemic significantly lower than young controls (p < 0.001).

2 Elderly anemic significantly lower than old controls (p < 0.02; p < 0.003; p < 0.001).

3 Elderly controls significantly less than young controls (p < 0.02).
no different in the elderly anemic than in the young or old controls. The mechanism for the erythropoietic findings is not clear. It seems likely that a defect in hematopoietic progenitor cell function exists. Although controversial, most studies indicate that the pluripotent hematopoietic stem cell (CFU-S) is minimally, if at all, decreased in aged animals.\textsuperscript{33,34} However, its ability to replicate when subjected to serial transfer may be altered.\textsuperscript{35,36} The fact that BFU-E levels were not reduced suggests that an absolute decrease in stem cell number did not account for the anemia. There clearly was, however, a defect in progenitor cell proliferation beginning at the CFU-E level. As the level of these progenitor cells and the more mature erythroid precursors is primarily regulated by Ep, a reduction in circulating Ep levels or a decreased cellular responsiveness to the hormone could account for the defect. This possibility is strengthened by our recent observation that the erythropoietic responsiveness to Ep in vivo and in vitro is reduced in the aged mouse.\textsuperscript{37} A second possibility is that a basic cellular abnormality exists. If this accounts for the lower number of progenitor cells measured, the mechanism remains to be determined. Although a basic defect in cellular proliferation may exist, multiple other cellular, biochemical, or hormonal alterations affecting progenitor cell proliferation in vitro must be also considered.

The presence of an alteration in progenitor cell function is made especially likely, as altered erythropoiesis was not the only defect found in elderly subjects with unexplained anemia. An overall reduction in hematopoiesis was present. This was reflected in lower peripheral leukocyte, neutrophil, and platelet count in the elderly with unexplained anemia, who also had significantly lower numbers of myeloid precursors than the elderly and young controls. In addition, a marked decrease in CFU-C levels was found.

A major question is whether or not these alterations result from the aging process or some other unrelated abnormalities. The values in the 15 elderly controls suggest that age may contribute to the defect as their bone marrow precursor measurements were generally intermediate between the young controls and the elderly with unexplained anemia.

Thus, this study demonstrates that the unexplained anemia in older subjects reflects a distinct abnormality. An overall reduction in hematopoiesis is present, the likely mechanism being an alteration in progenitor cell proliferation. The reduction in myelopoiesis is particularly relevant. It is well recognized that many aged subjects have impaired host defense mechanisms and a decreased ability to mount a neutrophil response to infection.\textsuperscript{38} It is likely that the presence of anemia may be an important marker of this abnormality and may identify those elderly individuals most likely to develop an inadequate response when stressed.

REFERENCES

Effect of age on hematopoiesis in man

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