A Correlative Study of the Erythrokinetics and Disturbances in Iron Metabolism Associated with the Anemia of Rheumatoid Arthritis

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THE ANEMIA of rheumatoid arthritis has attracted the attention of investigators for many years, but the pathogenesis of this anemia has remained elusive. All studies indicate that anemia is a frequent finding, and that it reflects the clinical activity of the disease. Disturbances in iron metabolism have also been noted. In all other respects the reported data are in marked disagreement. In the earlier work on this problem the importance of erythrocyte suppression as the basic mechanism in the pathogenesis of this anemia was usually stressed.1-5 These conclusions were reached chiefly on the basis of evaluation of the appearance of the bone marrow aspiration. These conclusions seem to have been confirmed with the use of radioactive iron studies in patients with infection and inflammation, although the subjects studied did not have rheumatoid arthritis.6-7 However, in a recent study the utilization of intravenously injected tracer doses of iron for hemoglobin synthesis was normal." Evidence for overt hemolysis in the anemia associated with infection or inflammation and specifically in the anemia of rheumatoid arthritis has been contradictory and not impressive.9

The most marked area of disagreement is in regard to the cause and relative importance of the disturbances in iron metabolism. Like most anemias associated with inflammation the anemia of rheumatoid arthritis is often hypochromic.2-8,9 Similarly, there is a moderate reduction in the serum iron level.8,9,14 The effect of iron therapy has been studied by several workers. Some have found that the hemoglobin levels improved with intravenous iron therapy, but this has been refuted by others.14,17 Examination of the bone marrow for available iron stores for hemoglobin production has yielded contradictory results, although in the majority of the studies marrow iron was found in at least some patients who were anemic.18-20 Very recently Freireich et al. have suggested that the disturbances in iron metabolism may be the result of a failure to mobilize iron from the reticuloendothelial cells.8,21

This study reports the simultaneous evaluation of erythrocyte production and destruction and disturbances in iron metabolism as measured by the most appropriate histologic, serologic and radioisotope technics.

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MATERIALS AND METHODS

Eighteen patients with active rheumatoid arthritis were selected for the erythrokinetic studies. Sixteen of these were male and two female. An additional six male patients were used for the radioactive iron oral absorption test. All of these patients were in relatively good nutritional state and were hospitalized at least during the initial part of the study. All were on regular hospital diets and received only conservative therapy consisting of bedrest, physiotherapy and aspirin. Patients were selected if the diagnosis of active rheumatoid arthritis was unquestioned, no attempt being made to select the more anemic patients. Patients who had a complicating illness or any evidence of blood loss were excluded from this study. An effort was made to study patients only when they had reached a relatively steady state of clinical activity. Complete blood counts were done in the usual manner and repeated at weekly intervals for the duration of the study. Stools were examined for the presence of occult blood three times prior to the initiation of study and once weekly thereafter. Bone marrow aspirations were done on all patients. Iron stores were evaluated in all instances on the bone marrow smears by the method of Rath and Finch. In many of the subjects, iron stores were further evaluated from fixed section preparations. The method of Kingsley and Getchell was used for determination of the serum iron. This test was repeated at least twice during the first month of study. The serum iron-binding capacity was done according to the method of Cartwright et al. and was also repeated twice during the first month.

Erythrokinetic studies were performed on each patient using Fe as a measurement of erythropoiesis and Cr as a measurement of erythrocyte destruction. These studies were carried out simultaneously in each subject. A preliminary report on the combined Fe-Cr radioisotope studies in patients with various hematologic disorders has been published previously. In this study these tests were performed in the following manner.

ERYTHROPOIETIC MEASUREMENTS. High specific activity Fe, 2 to 4.3 millicuries per milligram, was used in the chemical form of ferrous citrate. The patient was fasted on the morning of the first day of the study. Ten microcuries of Fe were added to approximately 20 milliliters of the patient's plasma and the mixture incubated for thirty minutes at 37 °C. The total quantity of inorganic iron injected ranged from 8 to 11 micrograms. An aliquot was saved for determination of the total radioactivity injected, and the plasma iron mixture was injected intravenously with the use of a weighed syringe. The rate of Fe disappearance from the plasma was determined by measuring the radioactivity in duplicate two milliliter plasma samples drawn at thirty minutes, one hour, two hours and three hours after the injection of the Fe. The sites of appearance of Fe in the body were determined by placing a directional scintillation counter without collimation over the sacrum as the site of bone marrow activity, the precordium as a measure of blood flow and over the liver and spleen areas. External surface counting was performed every hour for six hours during the first day, and daily for every other day subsequently for at least a two week period. Blood samples were drawn at least every other day for a period of two to three weeks for measurement of the utilization of the Fe by newly formed erythrocytes. The formulas for the plasma iron disappearance rates, plasma iron turnover rates and percent utilization of iron are the same as those previously published.

ERYTHROCYTE SURVIVAL MEASUREMENTS. High specific activity chromium was used in the chemical form of Na$_2$CrO$_4$. The labeling of the patient's blood with Cr was carried out on the third day of the study. At this time 100 to 150 microcuries of Cr were incubated with approximately 30 milliliters of blood for 30 minutes at 37 °C. The labeled blood was then washed twice with cold 0.9 per cent saline. The washed cells were resuspended in saline to approximately the original volume and an aliquot was removed to determine the total radioactivity injected. The labeled cells were then injected

*Reed-Curtis Scintistand. One inch lead shielded thallium-activated sodium iodide crystal, 1.5 inches x 1 inch.
intravenously with the use of a weighed syringe. A blood sample was taken in 30 minutes for the blood volume determination. Red cell life span was determined by measuring the radioactivity persisting in duplicate two milliliter hemolyzed whole blood samples withdrawn at appropriate intervals for approximately 30 days. No correction for Cr\textsuperscript{6+} elution was made. External surface counting for the appearance of Cr\textsuperscript{6+} in the body was performed in the manner as described for Fe\textsuperscript{5+}. The calculation of blood volume and red cell survival curves was done as published previously.\textsuperscript{16,17}

The gamma emissions of Cr\textsuperscript{6+} and Fe\textsuperscript{5+} in the blood and in the body were easily distinguished with the use of a single channel pulse height analyzer.\textsuperscript{18} With this instrument the radioactivity of Fe\textsuperscript{5+} was counted effectively at an energy level setting of 500 with an open channel. There was no contamination with Cr\textsuperscript{6+} radioactivity at this level. Cr\textsuperscript{6+} radioactivity was counted at an energy level of 210 with a channel width of 50. A small fraction of the radioactivity at the chromium energy level was due to emission of Fe\textsuperscript{5+}. This was corrected for in the following manner. An "iron factor" was determined prior to the injection of Cr\textsuperscript{6+} in the patient with the use of an Fe\textsuperscript{5+} standard:

\[
\text{Iron factor} = \frac{\text{c/s of Fe}\textsuperscript{5+} \text{ standard at chromium energy level}}{\text{c/s of Fe}\textsuperscript{5+} \text{ standard at iron energy level}}
\]

Actual Cr\textsuperscript{6+} radioactivity at chromium level =

\[
\text{Net c/s at chromium level} - (\text{iron factor} \times \text{net c/s Fe}\textsuperscript{5+} \text{ at iron level})
\]

The term "net counts" was used to indicate the actual count at each of the energy levels minus the background count, which for blood counting was room background and for body counting was the counts taken over the calf area. The use of a single channel pulse height analyzer for discriminating between the emissions of more than one isotope in human subjects has been substantiated by three recent reports.\textsuperscript{19-21}

SPECIAL FE\textsuperscript{5+} STUDIES. The oral iron absorption test using Fe\textsuperscript{5+} was performed in the following manner: The iron standard was prepared by dissolving 7.04 Gm. FeSO\textsubscript{4} (NH\textsubscript{4})\textsubscript{2} in 200 ml. of H\textsubscript{2}O, and adjusted to pH 2.4 with concentrated H\textsubscript{2}SO\textsubscript{4}. This preparation was stable for months at 4 C. For the performance of each test, 5 ml. of this standard was placed in a 100 ml. volumetric flask. 10 \mu c. of Fe\textsuperscript{5+} was added and distilled H\textsubscript{2}O added to final volume. 2 ml. aliquot was saved for counting, and the remaining solution was taken orally by the patient in a fasting state. The total amount of inorganic iron given to each patient was approximately 25 mg. Originally, blood samples were taken at one-half hour, one hour, one and one-half hours and two hours for determination of plasma activity. After it had been determined that the maximum plasma activity usually occurred at the one hour period, this time interval was used thereafter. A blood sample was taken in seven days for determination of the per cent utilization by erythrocytes. Plasma volume was determined by the use of the Cr\textsuperscript{6+} technic, and the peripheral blood hematocrit.

COUNTING PROCEDURE. (1) 1 ml. 50 per cent H\textsubscript{2}SO\textsubscript{4} and 10 ml. distilled H\textsubscript{2}O were added to a 2 ml. aliquot. Three ml. duplicate specimens were prepared for counting. (2) Duplicate 5 ml. plasma samples were prepared for counting from the 1 hour blood specimen. (3) Duplicate 6 ml. heparinized blood samples were prepared from the 7 day blood specimen. These were centrifuged and counted as packed red cells.

CALCULATIONS. (1) 1 Hour Plasma Activity:

\[
\text{Total counts injected} = \frac{98 \times C_1 \times 13}{2 \times 3}
\]

\[
\text{Per cent plasma appearance} = \frac{C_2 \times \text{P.V. (ml)} \times 100}{5 \times (\text{t.c.i.})}
\]

(2) 7 Day Red Cell Uptake = \[
\frac{C_3 \times \text{T.B.V. (ml)} \times 100}{6 \times (\text{t.c.i.})}
\]

\textsuperscript{*}Reed-Curtis Spectrometer, Model DAX-10, and Binary Scaler, CX14S-2.
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where \( C_1 \) = Counts/sec of 3 ml. Fe standard aliquot, \( C_2 \) = Counts/sec of 5 ml. plasma sample, and \( C_3 \) = Counts/sec of 6 ml. of 7 day sample; t.c.i. = total counts injected, P.V. = plasma volume, and T.B.V. = total blood volume.

The non-viable erythrocyte Fe\(^{59}\) incorporation study was performed according to the method of Freireich et al.\(^{30}\) The donor for this study was a patient with severe iron deficiency of a compatible blood group. The technique of Borum et al. was used to measure the Fe\(^{59}\) concentration in the top and bottom layers of erythrocytes from centrifuged blood.\(^{32}\)

**Results**

Table 1 and figures 1 and 2 summarize the hematologic results in our 18 patients with rheumatoid arthritis. It was clear in the early part of our study that our patients should be divided into two groups. Thirteen patients had typical rheumatoid arthritis, while five were officially classified as rheumatoid patients but were complicated in one or another respect. Two patients had what may be called malignant rheumatoid arthritis. Two had positive L.E. preparations, one of whom also had splenomegaly; one patient had splenomegaly with leukopenia as well as anemia. The patients in the atypical group were in general more severely ill. Despite the presence of positive L.E. preparations in two of these patients, the diagnosis of atypical rheumatoid arthritis rather than disseminated lupus erythematosus seemed justified on the basis of the over-all clinical and laboratory evaluation. The following points are worthy of specific mention: (1) Nine of the thirteen patients with typical rheumatoid arthritis had red cell volumes which were abnormally low. Four of the five atypical patients had abnormally low packed cell volumes. Although the range was great, the lowering of the packed cell volume was more marked in the atypical group. (2) Almost all of these patients had hypoferrremia, rapid disappearance of Fe\(^{59}\) from the plasma, and increased rates of Fe\(^{59}\) plasma turnover. (3) The per cent utilization of Fe\(^{59}\) by newly formed red cells was surprisingly good in both the typical and atypical groups. Only three of the 13 patients in the typical group had abnormally low utilization of Fe\(^{59}\) and in two of these the results were borderline. Two of the patients in the typical group had definitely abnormal Fe\(^{59}\) utilization. (4) Parenthetically, it should be added that the histologic examination of the bone marrow in these patients also failed to reveal any significant evidence of impaired erythropoiesis. The bone marrows of patients H. H. and W. F. showed a mild normoblastic hyperplasia. A mild to moderate degree of plasmacytosis and lymphocytosis was a common finding in most of the bone marrows. Marrow iron was present in all cases. In some patients, iron stores were modestly to moderately increased (patients 2, 7, 11, 13, 16, 18). (5) Six of the patients in the typical group and four of the patients in the atypical group had abnormal Cr\(^{51}\) red cell survival curves. It is to be emphasized that this degree of shortening of red cell life span cannot in itself account for the development of anemia in these patients since it should be appropriately compensated for by an uninhibited bone marrow.

Results of external surface monitoring for radioactive iron and chromium in these patients are summarized in figure 3. The majority of these patients had the findings as depicted in the patient D. E. There was rapid accumulation of radioactive iron over the sacrum followed by a normal release and the appearance of the radioactivity in the peripheral blood. The ratio of radio-
### Table 1.—Hematologic and Erythrokinetic Data on 18 Patients with Rheumatoid Arthritis

**Typical Rheumatoid Arthritis**

<table>
<thead>
<tr>
<th>Name</th>
<th>Age &amp; Sex</th>
<th>A.R.A. Class</th>
<th>Average Hgb. (gm.%)</th>
<th>Average P.C.V. (%g.)</th>
<th>Average M.C.C. (%)</th>
<th>Average Serum Trans (mg. %)</th>
<th>Average I.B.C. (mg. %)</th>
<th>Total Blood Volume (ml./kg.)</th>
<th>Red Cell Volume (ml./kg.)</th>
<th>Plasma Volume (ml./kg.)</th>
<th>Plasma Fr. Function (ml./kg.)</th>
<th>Plasma Fr. Turnover (mg./kg./day)</th>
<th>Feat. Occupation (mg./% day)</th>
<th>Cer. Survival (% of cells)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. H.W.</td>
<td>65</td>
<td>M</td>
<td>2</td>
<td>10.2</td>
<td>89</td>
<td>80.0</td>
<td>71</td>
<td>200</td>
<td>81.0</td>
<td>29.2</td>
<td>51.8</td>
<td>60</td>
<td>0.61</td>
<td>89/7</td>
</tr>
<tr>
<td>2. H.C.</td>
<td>21</td>
<td>M</td>
<td>2</td>
<td>12.1</td>
<td>42</td>
<td>30.0</td>
<td>97</td>
<td>200</td>
<td>68.2</td>
<td>27.9</td>
<td>40.3</td>
<td>40</td>
<td>0.99</td>
<td>78/15</td>
</tr>
<tr>
<td>3. J.S.</td>
<td>67</td>
<td>M</td>
<td>3</td>
<td>12.2</td>
<td>28</td>
<td>32.0</td>
<td>44</td>
<td>150</td>
<td>88.4</td>
<td>31.7</td>
<td>51.7</td>
<td>—</td>
<td>—</td>
<td>95/11</td>
</tr>
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<td>4. C.M.</td>
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<td>M</td>
<td>2</td>
<td>13.5</td>
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<td>31.0</td>
<td>88</td>
<td>284</td>
<td>66.8</td>
<td>29.7</td>
<td>57.1</td>
<td>37</td>
<td>0.58</td>
<td>79/12</td>
</tr>
<tr>
<td>5. F.K.</td>
<td>43</td>
<td>M</td>
<td>2</td>
<td>11.4</td>
<td>25</td>
<td>31.5</td>
<td>145</td>
<td>200</td>
<td>81.5</td>
<td>25.6</td>
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<td>55</td>
<td>1.47</td>
<td>82/7</td>
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<tr>
<td>6. C.W.</td>
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<td>M</td>
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<td>12.3</td>
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<td>31.0</td>
<td>118</td>
<td>200</td>
<td>81.1</td>
<td>27.1</td>
<td>56.0</td>
<td>57</td>
<td>0.75</td>
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<tr>
<td>7. D.E.</td>
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<td>3</td>
<td>12.1</td>
<td>29</td>
<td>31.3</td>
<td>128</td>
<td>300</td>
<td>64.5</td>
<td>25.0</td>
<td>59.5</td>
<td>23</td>
<td>2.18</td>
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<tr>
<td>8. J.B.</td>
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<td>M</td>
<td>2</td>
<td>12.6</td>
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<td>33.0</td>
<td>81</td>
<td>150</td>
<td>78.2</td>
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<td>44.6</td>
<td>30</td>
<td>1.23</td>
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<tr>
<td>9. F.W.</td>
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<td>M</td>
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<td>29.0</td>
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<td>300</td>
<td>83.0</td>
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<td>51.5</td>
<td>45</td>
<td>2.56</td>
<td>89/17</td>
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<tr>
<td>10. B.T.</td>
<td>30</td>
<td>M</td>
<td>3</td>
<td>10.5</td>
<td>24</td>
<td>30.0</td>
<td>116</td>
<td>300</td>
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<td>18.9</td>
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<td>45</td>
<td>1.21</td>
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<tr>
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<td>100/5</td>
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<tr>
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<td>46</td>
<td>M</td>
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<td>12.4</td>
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<td>81</td>
<td>200</td>
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<tr>
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<td></td>
<td></td>
<td>12.2</td>
<td>38</td>
<td>31.2</td>
<td>110</td>
<td>230</td>
<td>74.6</td>
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<td>46.7</td>
<td>43</td>
<td>1.18</td>
<td>87/11</td>
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</table>
## Atypical Rheumatoid Arthritis

<table>
<thead>
<tr>
<th>Name</th>
<th>Age &amp; Sex</th>
<th>A.R.A. Class</th>
<th>Average Hgb. (Gm./%)</th>
<th>Average P.C.V.</th>
<th>Average M.C.H. C. (%)</th>
<th>Average M.C.H. C. (%)</th>
<th>Average I.E.C. (g/dl.)</th>
<th>Total Blood Volume (ml./Kg.)</th>
<th>Red Cell Volume (ml./Kg.)</th>
<th>Plasma Volume (ml./Kg.)</th>
<th>Plasma Fibrinolytic Activity</th>
<th>Fe-S turnover (mg. K/kg/day)</th>
<th>Fe absorption (mg. K/kg/day)</th>
<th>Cell Survival (T% days)</th>
<th>Complication</th>
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<tbody>
<tr>
<td>Normal Values</td>
<td>M</td>
<td>14-16</td>
<td>12-16</td>
<td>40-54</td>
<td>37-47</td>
<td>22-36</td>
<td>12-15</td>
<td>160-200</td>
<td>200-250</td>
<td>25-32</td>
<td>35-45</td>
<td>60-90</td>
<td>0.46-0.72</td>
<td>80-90</td>
<td>In 7-12</td>
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<tr>
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<td>48</td>
<td>M</td>
<td>4</td>
<td>12.0</td>
<td>11.0</td>
<td>96</td>
<td>145</td>
<td>54.0</td>
<td>17.0</td>
<td>37.0</td>
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<td>1.30</td>
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<td>M</td>
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<td>137</td>
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<td>85.1</td>
<td>55.0</td>
<td>50.0</td>
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<td>91/11</td>
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<td>73.6</td>
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<td>1.60</td>
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<td>33.0</td>
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<td>34</td>
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<td>61</td>
<td>200</td>
<td>56.0</td>
<td>21.0</td>
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<td>64/11</td>
<td>25.5</td>
<td>Malignant R.A.</td>
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<td>18. G.G.</td>
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<td>91</td>
<td>200</td>
<td>70.5</td>
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<td>46.8</td>
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<td>1.68</td>
<td>81/11</td>
<td>23.0</td>
<td>Neutropenia</td>
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activity over the liver as compared to the spleen was normal (1.2:1). External surface monitoring for the radioactive chromium revealed a gradual decline of the radioactivity in the blood with no abnormal accumulation over the spleen. These findings must be considered normal. An occasional patient, however, did have evidence of abnormal erythrokinetics as was seen in patient A. N., who is a member of the atypical group. There was a less than normal accumulation of radioactive iron over the sacrum, with an abnormal accumulation over the liver. This was evidence of functional impairment of erythropoiesis corresponding to the abnormal \( \text{Fe}^{59} \) incorporation in the blood of 64 per cent (table 1 and fig. 1). Patient G. G. is an example of a patient having an abnormality in external surface monitoring with an enlarged spleen secondary to the rheumatoid process. The abnormality to be noted is the reversal of the liver to spleen ratio in the external monitoring for both \( \text{Fe}^{59} \) and \( \text{Cr}^{51} \). This was evidence of splenic sequestration associated with the splenomegaly. These findings were confirmed by the results of the blood survival curve in this patient, which indicated a moderately severe hemolytic anemia (table 1 and fig. 2). This patient was splenectomized because of the severe pancytopenia which was thought to be due to a hypersplenic state. This patient expired several days postoperatively from an overwhelming staphylococcal infection of the lungs. Histologically, the spleen revealed only the findings consistent with congestive splenomegaly.
ANEMIA OF RHEUMATOID ARTHRITIS

Our findings regarding the severity of anemia in rheumatoid arthritis are graphically depicted in figure 4. Nine of the 18 patients had plasma volume determinations which were greater than normal. Eight of these nine patients had peripheral blood hematocrits which were below forty volumes per cent. However, only three would be considered anemic if they were plotted according to their actual red cell volume as determined by the Cr\textsuperscript{51} method. These findings indicate that in some instances the so-called anemia of uncomplicated rheumatoid arthritis may be a phenomenon of plasma volume dilution.

Figure 5 illustrates that there was a relationship between the lowering of the serum iron level and the anemia in patients with rheumatoid arthritis, whether that anemia was measured by the peripheral hematocrit or the actual determination of red cell volume. Those patients with the lowest red cell volumes or the lowest packed volumes had the lowest serum irons. It may be inferred, then, the hypoferremia was related to the development of the anemia in rheumatoid arthritis. How may it have been related?

Table 2 summarizes the findings regarding absorption of iron in patients with rheumatoid arthritis as compared with normals and patients with documented iron deficiency due to blood loss. The difference between the absorption of radioactive iron from the gastrointestinal tract in rheumatoid arthritis patients as compared to normals could not be considered significant under the conditions of this procedure. This was true whether one measured the per
Fig. 3.—Combined Fe\(^{59}\)-Cr\(^{51}\) external monitoring in patients with rheumatoid arthritis. Patient D. E. reveals normal findings. Patient A. M. shows dyserythropoiesis characterized by an abnormal accumulation of Fe\(^{59}\) over the liver associated with a less than normal appearance of Fe\(^{59}\) over the sacrum. Typical findings of splenic sequestration in a rheumatoid patient with splenomegaly are seen in the external monitoring of patient G. G.
cent absorbed in one hour or the per cent appearing in red cells in seven days. Despite the hypoferremia which was present in each of the six patients with rheumatoid arthritis there appeared to be no stimulus to increased absorption of iron from the gastrointestinal tract such as was seen in the six patients with iron deficiency.

As mentioned above, Freireich et al. have presented evidence that the hypo-
ferremia associated with the anemia of inflammation may be the result of a failure to mobilize iron from the reticuloendothelial cells. The major source of this iron in the reticuloendothelial cells would be iron broken down from senescent red cells. If such a defect exists in patients with rheumatoid arthritis, it might be detected by employing the technic recently reported by Borun et al. for the measurement of the mean life span of erythrocytes labeled with Fe-59. Using those authors’ technic we measured the relative concentrations of Fe-59 in the top and bottom layers of erythrocytes over a period of at least 150 days. These results in six patients with rheumatoid arthritis are shown in figure 6. The findings are very similar to those reported in normals by Borun et al. Parenthetically, it was of great interest that patients H. H. and W. F. showed an earlier decline of the radioactivity from the bottom layer as the cells aged. One explanation for this might involve accelerated senescence of erythrocytes in these patients, and happened to correlate well with the presence of anemia and accelerated red cell destruction in these patients (table 1).

The exchange of radioactive iron between the layers of a centrifuged tube probably could not be considered a very sensitive test to detect a defect in the mobilization of iron. For this reason we employed the method of Freireich et al. in an attempt to demonstrate the possible presence of such a defect. Senescent red cells were prepared in the manner suggested by Freireich. The cells were taken from an iron-deficient donor of the compatible blood group. An additional precaution regarding the non-viability of these erythrocytes was used: We measured the glycolysis of these non-viable red cells after the appropriate incubation and found it to be completely suppressed. These radioactive non-viable red cells were then injected into patient W. R., a rheumatoid arthritic, as well as a normal control. The rate of incorporation of these

| Table 2.—Absorption of Fe-59 from G.I. Tract and Subsequent Incorporation into Red Cells |
|---------------------------------|---------------------------------|
| **Group**                      | **Per Cent Absorption 1 Hour** | **Per Cent Incorporation in Red Cells in 7 Days** |
| Normal (6)                     | 0.57 (0.12 – 1.71)             | 2.06 (0.28 – 4.59) |
| Rheumatoid Arthritis (6)      | 1.17 (0.03 – 3.11)             | 3.83 (0.59 – 7.15) |
| Iron Deficiency (6)            | 7.50 (1.54 – 12.1)             | 30.9 (11.4 – 54.4) |
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FIG. 6.—Fe concentration in top and bottom layers of erythrocytes from centrifuged blood. It is to be noted that there appears to be no impairment in the exchange of iron between the upper and bottom layers, indicating a normal reutilization of iron from senescent red cells. Patients H. H. and W. F. show a decline in the radioactivity of the bottom layer beginning earlier than normal. This suggests accelerated senescence.

Senescent red cells in the patient with rheumatoid arthritis as compared to the control is seen in figure 7. There was a suggestive delay in the appearance of the radioactivity in the patient as compared to the control, but the final percentage incorporation was not impressively different. Following the completion of these studies both the patient and control were studied with intravenously injected radioactive iron incubated in their own plasma; these findings were normal in both subjects.

Discussion

Our findings reported in this study confirm the well known fact that the anemia of rheumatoid arthritis reflects the activity of the disease. We are in complete agreement with the findings of moderate degrees of hypoferremia in most cases as reported by previous investigators. A slight degree of hypochromia was usually noted in the peripheral blood smear of these patients.
However, calculation of the mean corpuscular hemoglobin concentration revealed no significant deviation from the normal (table 1). We agree with Freireich et al., Coons et al., and McCrea that the anemia of rheumatoid arthritis is not an iron deficiency anemia in the usual sense. Our patients had normal to low iron-binding capacities, normal absorption of iron from the gastrointestinal tract and ample to increased iron stores in the bone marrow. Further, we found that the parenteral injection of moderate amounts of iron did not result in any significant reticulocytosis or improvement in the hemoglobin level in a few of these patients in whom this form of therapy was tried.

The increased plasma volumes found in some patients with active rheumatoid arthritis is in agreement with the work of Dixon et al. However, we are reluctant to attribute the anemia of uncomplicated rheumatoid arthritis entirely to increased plasma volume for the following reasons: First, we did not measure the plasma volume directly. The blood volume determinations in this study were measured with Cr which is a red cell tag, and the plasma volume results reported here are based on a calculation using the peripheral blood hematocrit. On the other hand, Dixon et al. did measure the plasma volume directly, using Evans blue dye with the same results as ours. Secondly, we demonstrated other abnormalities in erythrocyte metabolism, namely, shortening of red cell survival and abnormalities in iron metabolism. These
abnormalities must play a part in the pathogenesis of the anemia in rheumatoid arthritis. We have no explanation for the elevation of plasma volume found in this study.

Our results indicate that patients with rheumatoid arthritis utilize intravenously injected radioiron for erythrocyte production in a normal manner. The actual amount of iron used per day for hemoglobin synthesis was either normal or greater than normal except in patient 17, in whom it was slightly decreased. This statement is based on the results of the plasma iron turnover rates and the per cent iron utilization as tabulated in Table 1. The milligrams of iron available for hemoglobin formation per day varied from 19.8 (patient 17) to 158.3 (patient 15), with a mean of 71.8. Furthermore, we found no impairment of erythropoiesis as evaluated by means of bone marrow aspirations.

The results of the radiochromium red cell survival studies in our patients indicate that hyperhemolysis contributes to the anemia in rheumatoid arthritis. Patients with atypical rheumatoid arthritis tended to have a greater degree of red cell destruction than those patients with a more classic form of this disease. It is to be emphasized again that the magnitude of increased red cell destruction found in these patients cannot account for the development of anemia, inasmuch as their bone marrows should adequately compensate for this degree of impairment. The fact that some of these patients do get anemic must mean that there is an inappropriate bone marrow response. Freireich’s hypothesis regarding a defect in the mobilization of iron might be the explanation for this inadequate bone marrow response.8,21 We have attempted to extend this hypothesis to the human subject, but our results to date are inconclusive. There is at least a suggestion of a defect in the availability of iron from senescent red cells in the results herein reported, and further studies are in progress.

It is of interest that our patients in general are not as anemic as those reported by other workers.8,9,16,29 There are at least two possible explanations for this. One is that most of our patients were male subjects. Rheumatoid arthritis is more frequent in women, and our clinical impression has been that the anemia tends to be somewhat more severe in the female. Further, women in the younger age group might well have a greater tendency to develop anemia, because of their marginal iron stores. The second explanation is that our patients were hospitalized for at least the greater part of these experiments and were not started on the study until some degree of clinical stability had been achieved. At times this interval was as long as two months, during which time the patient had the potential benefits of a favorable hospital environment and a well supervised diet. It is a well established fact that hospitalization, bed rest and a good diet often result in an amelioration of the rheumatoid processes including the anemia.

We have been impressed with the diagnostic importance of a severe degree of anemia in patients with rheumatoid arthritis. Although this is certainly consistent with the rheumatoid state, in our opinion it always suggests the possibility of complications. These include superimposed iron deficiency due
to blood loss, the development of the malignant phase of rheumatoid arthritis, the presence of congestive splenomegaly, or the presence of underlying disseminated lupus erythematosus rather than rheumatoid arthritis.

CONCLUSIONS

The anemia of rheumatoid arthritis has been investigated in 18 patients, 16 of whom were males, under favorable hospital conditions during a period of relative clinical stability. Six other male patients with active rheumatoid arthritis were used to measure the absorption of radioactive iron from the gastrointestinal tract. Our findings indicate that the anemia of uncomplicated rheumatoid arthritis is modest under the conditions of this study. In some instances the anemia may be due at least in part to plasma dilution. There was no evidence of iron deficiency in any of these patients. We detected no histologic evidence of suppressed erythropoiesis. Functional impairment of erythropoiesis was found in five patients. Impairment of red cell survival was found in ten patients. This was more marked in the patients having enlarged spleens. The magnitude of impairment in red cell survival found in this study cannot in itself account for the development of anemia. The inability of patients with rheumatoid arthritis to adjust to this impairment of erythrocyte survival may be due to a defect in mobilization of iron from the reticuloendothelial cells. Our studies to date cannot confirm or refute this hypothesis. We feel that a severe anemia associated with rheumatoid arthritis suggests complications, such as superimposed iron deficiency, malignant rheumatoid disease, hypersplenism, or an underlying diagnosis of disseminated lupus erythematosus.

SUMMARIO IN INTERLINGUA

Le anemia de arthritis rheumatoide esseva investigate in 18 patientes—incluse 16 masculos—sub favorabile conditiones hospitalari durante un periodo de relative stabilitate clinica. Sex altere patientes de sexo masculin con arthritis rheumatoide esseva usate pro mesurar le absorption de ferro radioactive ab le vias gastrointestinal. Nostre datos indica que le anemia de non-complicate arthritis rheumatoide es modeste sub le conditiones que prevaleva in le presente studio. In certe casos le anemia es posibilemente le effecto—al minus in parte—de dilution de plasma. Nulle signo de carentia de ferro esseva notate in ulle del patientes del presente serie. Esseva trovate nulle indicio de un suppression del erythropoiesis. Dysfunctions erythropoietic esseva trovate in cinque patientes. Reducite periodos de superviventia erythrocytic esseva constatate in dece patientes. Iste observation esseva le plus frappante in patientes con allargamento del splen. Le mesura del reduction del supervivencia erythrocytic trovate in iste studio non pote, per se, explicar le disveloppamento de anemia. Le incapacitate de patientes con arthritis rheumatoide de adjustar se a iste reducite supervivencia erythrocytic es posibilemente le consequentia de un defecto in le mobilisation de ferro ab le cellulas reticuloendothelial. Le studios effectuate usque al tempore presente non pote confirmar o refutar le hypothese mentionate. Nos opinia que un anemia sever que occurre in association con arthritis rheumatoide suggere que complicaciones es presente, per exemplo un superimponite carentia de
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ferro, maligne morbo rheumatoide, hypersplenismo, o un subjacente syndrome de disseminate lupus erythematoso.

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A Correlative Study of the Erythrokinetics and Disturbances in Iron Metabolism Associated with the Anemia of Rheumatoid Arthritis

IRWIN M. WEINSTEIN and Donna Mobley