Plasma lactate dehydrogenase in megaloblastic anaemia

C. F. McCarthy, I. D. Fraser, and A. E. Read

From the Departments of Medicine and Haematology, Royal Infirmary, Bristol

SYNOPSIS
Raised plasma lactate dehydrogenase (LDH) values were found in 26 patients with marked megaloblastic anaemia due either to vitamin B₁₂ or folic-acid deficiency or a combination of these factors.

Minor megaloblastic changes were not usually accompanied by plasma LDH elevation. Serial LDH estimations were as valuable as serial reticulocyte counts in assessing the response to physiological doses of folic acid and therefore in deciding whether megaloblastic anaemia is partially or completely due to folic-acid deficiency.

Elevation of plasma lactate dehydrogenase concentration (LDH) is known to occur in patients with megaloblastic anaemia. The literature on this subject is reviewed by Hess (1963) and the value of LDH estimations as a screening test in the detection of megaloblastic anaemia during pregnancy is commented on by Fleming and Elliott (1964).

Our studies were carried out to provide further information on LDH levels in megaloblastic anaemia and to evaluate the usefulness of serial estimations in assessing the response to physiological amounts of folic acid, and thus as a test of folic-acid deficiency.

PATIENTS AND METHODS
Three-hundred and seventy-three LDH estimations were carried out on the following groups of patients:
Twenty-seven patients whose bone marrow showed marked megaloblastic changes; 10 patients whose marrow showed minimal megaloblastic or megaloblastoid change;
27 post-gastrectomy patients with and without anaemia;
16 patients with miscellaneous conditions, mainly peptic ulcer or neuritis, who served as a control group.
In 13 of the patients with megaloblastic anaemia the haematological response to 50 μg of folic acid daily was studied. Nine of these had predominant vitamin B₁₂ deficiency, four folic-acid deficiency. The folic acid was given intramuscularly to the patients with malabsorption and orally to the others. Reticulocytes and plasma LDH were estimated daily, haemoglobin and packed-cell volume on alternate days. When steady base-line values for reticulocytes were obtained the folic acid was started and continued for 10 days except where otherwise stated in the test. These patients were on a white diet containing insufficient folic acid or B₁₂ to cause a reticulocyte response during the period of folic-acid administration.

URINARY FORMININO-GLUTAMIC ACID (FIGLU) An oral loading dose of 15 g of L-histidine was given and the urinary excretion of Figlu was measured by conventional voltage electrophoresis on cellulose acetate (Kohn, Mollin, and Rosenbach, 1961). Although the method is not quantitative a rough grading of the Figlu spot was made: + + + +. It has been shown that + + + represents a urinary Figlu concentration greater than 500 μg/ml.

Serum folate levels were measured by microbiological assay (L. casei ATCC 746), the normal range being 5-9 to 21 μg/ml. Serum vitamin B₁₂ levels were measured by microbiological assay (L. leichmanii), the normal range being 140 to 900 μg/ml.

Lactate dehydrogenase was estimated as described by Berger and Brodia (1964). The normal value quoted by these authors is 100 to 350 units per ml., 350 to 500 being borderline, and above this abnormal. As some of our control group had values up to 500 units per ml we have taken this figure as the upper limit of normal.

Marrow reassessment The initial marrow films in 10 patients (Table I) were reported to show mild megaloblastic or megaloblastoid changes. These marrow films were re-examined by one of us and a rough grading of the degree of megaloblastic change present was made. For this purpose the red and white cell precursors were separated and the grading was carried out as follows:
Megaloblastic changes present at all stages of red cell development + + + ; changes present in only a few intermediate and late normoblasts +, changes intermediate between + and +++ were designated ++. When numerous macropolyocytes and giant metamyelocytes were present the white cell series was designated ++++, when only a few macrocytophytes and giant metamyelo-
cytes were present +, and intermediate changes ++.

Received for publication 24 September 1965.

1 Present address: Cheltenham General Hospital.

51
C. F. McCarthy, I. D. Fraser, and A. E. Read

The term megaloblastoid (M) was used to describe the marrow appearance when red cell precursors showed slight asynchronism between nuclear and cytoplasmic maturation, no Howell Jolly bodies were present, and the white cell series was normal.

RESULTS

The LDH values of patients whose marrows showed marked megaloblastic changes are shown in Figure 1. All values were above the normal range.

The haematological data and the results of reassessment of the marrow picture, together with the type of treatment of the 10 patients whose marrow was initially reported as showing mild megaloblastic or megaloblastoid change, are shown in Table I.

Two patients had LDH values greater than normal. Case 1 had post-gastrectomy vitamin B12 deficiency and required vitamin B12 before the haemoglobin level rose. Case 3 was a fit 47-year-old man who had persistently elevated levels although significant vitamin B12 or folic acid deficiency was not present. His haemoglobin rose rapidly when intravenous iron was given and LDH fell to normal levels.

EFFECTS OF TREATMENT WITH PHYSIOLOGICAL AMOUNTS OF FOLIC ACID ON LDH VALUES IN PATIENTS WITH MEGALOBLASTIC ANAEMIA. Details of patients with predominant vitamin B12 deficiency are given in Table II. In six patients (1 to 6) there was no increase in haemoglobin, packed cell volume, reticulocyte count, nor was there any significant change in LDH level during the 10-day period of treatment with physiological doses of folic acid. One patient (no. 7) developed a

10% increase in reticulocyte count without a fall in LDH level but she was unusual in that her reticulocyte count on admission to hospital was 7% and the serum contained large amounts of methaemalbumin. Over a few days the reticulocyte count fell to a base line value of 3 to 4% without a change in the methaemalbumin concentration, but with a 5% fall in haemoglobin level. The folate level and Figlewicz excretion in this patient did not suggest folic acid deficiency.

In two patients (cases 8 and 9) considerable alteration in LDH occurred associated with reticulocyte changes. In case 8 the LDH fell from a pretreatment level of 4,000 units to 2,000 and this fall

| TABLE I |

<table>
<thead>
<tr>
<th>CASE NO.</th>
<th>MEGALOBLASTIC CHANGE IN MARROW</th>
<th>Hb (g./100 ml.)</th>
<th>TREATMENT</th>
<th>LDH (U. units/ml.)</th>
<th>Serum B12 (t.ug./ml.)</th>
<th>Serum Folate (mg./ml.)</th>
<th>Serum Iron (mg./ml.)</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>+ +</td>
<td>67</td>
<td>99</td>
<td>Vitamin B12</td>
<td>750</td>
<td>118</td>
<td>4-6</td>
<td>150</td>
</tr>
<tr>
<td>2</td>
<td>0</td>
<td>65</td>
<td>89</td>
<td>Penicillin</td>
<td>420</td>
<td>143</td>
<td>1-4</td>
<td>—</td>
</tr>
<tr>
<td>3</td>
<td>Megaloblastoid</td>
<td>48</td>
<td>91</td>
<td>Intravenous iron</td>
<td>1,750</td>
<td>270</td>
<td>6-5</td>
<td>16</td>
</tr>
<tr>
<td>4</td>
<td>+</td>
<td>66</td>
<td>78</td>
<td>Folic acid</td>
<td>360</td>
<td>300</td>
<td>1-4</td>
<td>111</td>
</tr>
<tr>
<td>5</td>
<td>+</td>
<td>76</td>
<td>100</td>
<td>Intravenous iron</td>
<td>260</td>
<td>260</td>
<td>6-2</td>
<td>67</td>
</tr>
<tr>
<td>6</td>
<td>Megaloblastoid</td>
<td>78</td>
<td>101</td>
<td>Intravenous iron</td>
<td>290</td>
<td>140</td>
<td>1-3</td>
<td>50</td>
</tr>
<tr>
<td>7</td>
<td>+</td>
<td>57</td>
<td>79</td>
<td>Iron, vitamin B12</td>
<td>260</td>
<td>280</td>
<td>2-3</td>
<td>62</td>
</tr>
<tr>
<td>8</td>
<td>+</td>
<td>83</td>
<td>84</td>
<td>Folic acid</td>
<td>200</td>
<td>200</td>
<td>4-7</td>
<td>—</td>
</tr>
<tr>
<td>9</td>
<td>Megaloblastoid</td>
<td>48</td>
<td>93</td>
<td>Intravenous iron</td>
<td>290</td>
<td>269</td>
<td>4-4</td>
<td>—</td>
</tr>
<tr>
<td>10</td>
<td>Megaloblastoid</td>
<td>77</td>
<td>88</td>
<td>Oral iron</td>
<td>260</td>
<td>253</td>
<td>2-0</td>
<td>40</td>
</tr>
</tbody>
</table>

FIG. 1 Plasma LDH levels in patients with markedly megaloblastic bone marrow (14-6 g. = 100).
Plasma lactate dehydrogenase in megaloblastic anaemia

TABLE II
PATIENTS WITH VITAMIN B12 DEFICIENCY

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Haemoglobin (g./100 ml.)</th>
<th>LDH Before Treatment</th>
<th>LDH After Treatment</th>
<th>Serum Vitamin B12 (µg./ml.)</th>
<th>Serum Folate (mg./ml.)</th>
<th>Figlu</th>
<th>Schilling Test (%)</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>55</td>
<td>8,000</td>
<td>8,700</td>
<td>10</td>
<td>3.4</td>
<td>-ve</td>
<td>1.32</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>2</td>
<td>53</td>
<td>5,050</td>
<td>6,400</td>
<td>75</td>
<td>5.8</td>
<td>-ve</td>
<td>1.7</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>3</td>
<td>47</td>
<td>3,750</td>
<td>4,200</td>
<td>70</td>
<td>20.9</td>
<td>-ve</td>
<td>0.14</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>4</td>
<td>40</td>
<td>1,650</td>
<td>1,700</td>
<td>57</td>
<td>2.6</td>
<td>+</td>
<td>0.14</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>5</td>
<td>48</td>
<td>2,250</td>
<td>1,850</td>
<td>50</td>
<td>3.5</td>
<td>++</td>
<td>0.2</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>6</td>
<td>60</td>
<td>2,200</td>
<td>3,200</td>
<td>94</td>
<td>3.5</td>
<td>+</td>
<td>4</td>
<td>Post-gastrectomy</td>
</tr>
<tr>
<td>7</td>
<td>40</td>
<td>12,500</td>
<td>17,500</td>
<td>110</td>
<td>8.5</td>
<td>+</td>
<td>0.9</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>8</td>
<td>35</td>
<td>4,000</td>
<td>2,000</td>
<td>95</td>
<td>8.2</td>
<td>++++</td>
<td>0.5</td>
<td>Pernicious anaemia</td>
</tr>
<tr>
<td>9</td>
<td>40</td>
<td>10,000</td>
<td>5,200</td>
<td>104</td>
<td>2.7</td>
<td>++++</td>
<td>1.2</td>
<td>Pernicious anaemia</td>
</tr>
</tbody>
</table>

*Ten days' oral folic acid, 50 µg. a day.

TABLE III
PATIENTS WITH FOLIC ACID DEFICIENCY

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Haemoglobin (g./100 ml.)</th>
<th>LDH Before Treatment</th>
<th>LDH After Treatment</th>
<th>Serum Folate (mg./ml.)</th>
<th>Figlu</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>42</td>
<td>3,900</td>
<td>500</td>
<td>95</td>
<td>2.7</td>
<td>++++</td>
</tr>
<tr>
<td>2</td>
<td>58</td>
<td>720</td>
<td>390</td>
<td>260</td>
<td>1.8</td>
<td>nd</td>
</tr>
<tr>
<td>3</td>
<td>42</td>
<td>1,290</td>
<td>520</td>
<td>190</td>
<td>1.2</td>
<td>nd</td>
</tr>
<tr>
<td>4</td>
<td>35</td>
<td>15,000</td>
<td>4,000</td>
<td>210</td>
<td>1.4</td>
<td>+++</td>
</tr>
</tbody>
</table>

was associated with a 14% increase in reticulocytes. By the twelfth day the reticulocyte count had fallen to baseline levels. Three further LDH values at this time were approximately 2,000. No alteration of haemoglobin level occurred during these 12 days. In case 9 a similar response occurred during the first 10 days; the LDH values fell by just under 5,000 units in association with a reticulocytosis. The LDH values were followed for a further 20 days during which folic acid in physiological doses was continued. During this time the LDH value rose gradually to the pre-treatment value of 10,000. The haemoglobin level did not rise during the period of observation. These two patients had strongly positive Figlu tests and were considered to have combined deficiency of vitamin B12 and folic acid. All of these patients then responded promptly to vitamin B12 and the LDH values fell to normal.

**DISCUSSION**

In common with the findings of others the LDH levels in our patients with marked megaloblastic changes in their bone marrow were considerably raised above normal. Patients with slight megaloblastic changes usually had normal levels. This latter

**TABLE IV**
SERUM FOLATE, VITAMIN B12, AND LDH VALUES IN NON-MEGALOBLASTIC PATIENTS

<table>
<thead>
<tr>
<th>Serum Folate (mg./ml.)</th>
<th>Serum Vitamin B12 (µg./ml.)</th>
<th>No. of patients</th>
<th>LDH ± ISD</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt; 4</td>
<td>&lt; 4</td>
<td>29</td>
<td>404 ± 183</td>
</tr>
<tr>
<td>420 ± 173</td>
<td>404 ± 185</td>
<td>13</td>
<td>397 ± 170</td>
</tr>
<tr>
<td>36</td>
<td>7</td>
<td>150</td>
<td>&lt; 150</td>
</tr>
</tbody>
</table>

**TABLE V**
SERUM FOLATE, VITAMIN B12, AND LDH IN PATIENTS AFTER PARTIAL GASTRECTOMY

<table>
<thead>
<tr>
<th>Serum Vitamin B12 (µg./ml.)</th>
<th>No. of patients</th>
<th>LDH ± ISD</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt; 500</td>
<td>7</td>
<td>266 ± 131</td>
</tr>
<tr>
<td>&lt; 500</td>
<td>20</td>
<td>273 ± 88</td>
</tr>
<tr>
<td>10</td>
<td>Serum folate (mg./ml.)</td>
<td>5.3 ± 2.7</td>
</tr>
<tr>
<td>19</td>
<td></td>
<td>6.1 ± 2.7</td>
</tr>
</tbody>
</table>
finding might be expected when it is considered that
the LDH originates in the megaloblasts of the bone
marrow and then passes to the peripheral blood
(Elliott and Fleming, 1965). The LDH produced by a
few megaloblastic cells might not be adequate to
raise the peripheral LDH levels. Studies on the bone
marrow LDH might have been more rewarding in
these patients as elevation of LDH levels would first
be detectable here. From our results it may be
concluded that LDH estimation does not offer a
simple means of detecting minor megaloblastic
changes but minor megaloblastic change may not be
significant, particularly in the presence of iron
deficiency.

The decision as to whether megaloblastic anaemia
is due to folic acid or vitamin B₁₂ deficiency is some-
times difficult. In such instances it may be necessary
to carry out a therapeutic trial with physiological
amounts of folic acid and to assess response by
means of daily reticulocyte counts. Daily LDH
estimations would seem to be at least as valuable as
reticulocyte counts in assessing the response to a
therapeutic trial of folic acid. In two patients, cases
8 and 9, who had combined vitamin B₁₂ and folic acid
deficiency, a reticulocyte response was accompanied
by a fall in LDH value. The LDH values in these
patients did not, however, fall to normal, as they
would if folic acid deficiency alone was present.
From the partial return towards normal of LDH
values it was possible to diagnose the combined
deficiency which could not be detected from the
reticulocyte response alone. In one patient (case 7),
who did not appear to have an associated folic acid
deficiency, a reticulocyte response occurred which
was not accompanied by a fall in LDH so perhaps in
this patient the LDH was superior to the reticulocyte
count in assessing the response. The six patients with
vitamin B₁₂ deficiency alone showed no depression of
LDH or increase in reticulocyte count during the
period of the trial whereas the patients with folic acid
deficiency showed a rapid fall towards normal.

From Tables IV and V it is clear that low serum
levels of vitamin B₁₂ or folic acid are not of them-
selves accompanied by elevated LDH values nor are
the few LDH values above 500 in normal patients
accompanied by altered B₁₂ or folic acid levels.

We wish to thank Dr. J. R. Clamp and Dr. A. B. Raper
for their help in this study and their advice with the writing
of this report.

REFERENCES

Plasma lactate dehydrogenase in megaloblastic anaemia

C. F. McCarthy, I. D. Fraser and A. E. Read

*J Clin Pathol* 1966 19: 51-54
doi: 10.1136/jcp.19.1.51

Updated information and services can be found at:
http://jcp.bmj.com/content/19/1/51

**Email alerting service**

*These include:*

Receive free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

**Notes**

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/