Correlation of Lactate Dehydrogenase in Megaloblastic Anemia.

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Abstract:
Hematology is a rapidly changing discipline. It has experienced a dramatic shift from dependence on a simple lab observation into the field of basic science. The diagnosis of blood disorders will continue to require correlation of both clinical and lab evidences. The laboratory has major role in providing screening services and provisional diagnosis of blood disorders which may be studied and investigated further in much more detail.
This was a cross sectional study and was carried out in the Department of Pathology, MGM medical college and hospital, Aurangabad [MS], India during period from Oct. 2013 to Oct. 2015 in 50 patients in each Group of cases & Control.
In present study it was observed that majority of patients of megaloblastic anemia i.e 78% had predominantly macrocytic anemia and 76% Cases of megaloblastic anemia had hypersegmented neutrophils in peripheral blood smear examination. Mean serum LDH calculated in this study was 2396.04 whereas it was 316.3 in control group. The above table depicts that there was marked increase in serum LDH up to 5 times of the upper normal limit of serum LDH which was statically significant with p value (0.000). Thus we conclude that serum LDH may have an important role in diagnosis of megaloblastic anemia.

Key Words: Megaloblastic Aneamia, Serum Lactate Dehydrogenase, Hemoglobin etc.

Introduction:
Man since time immemorial has been plagued by diseases and man since the same time has been in turn fascinated and interested in these diseases.
Hematology is a rapidly changing discipline. It has experienced a dramatic shift from dependence on a simple lab observation into the field of basic science. The diagnosis of blood disorders will continue to require correlation of both clinical and lab evidences. The laboratory has major role in providing screening services and provisional diagnosis of blood disorders which may be studied and investigated further in much more detail.
Anemia constitutes most important part of hematological disorders which may require early diagnosis and treatment. Of various anemias, nutritional anemias are more common in our country which may be due to iron, Vit. B12 or folate deficiency. The term “Megaloblast” was first coined by Ehrlich [1], which shows characteristic “Sieve” like nuclear chromatin with abundant basophilic cytoplasm.
Megaloblastic anemia constitutes a considerable health problem in developing countries like India. The incidence of megaloblastic anemia in various part of our country is ranging from 3.1 to 71.1%.
Macrocytes, macro-ovalocytes and hypersegmented neutrophils on peripheral blood smear provide supportive evidence in the diagnosis of megaloblastic anemia. But for the confirmation of diagnosis of megaloblastic anemia bone marrow examination is required which is invasive procedure and may not be available at peripheral centers.

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Besides hematological investigations certain biochemical parameters like serum Vit.B12 serum folate, radioactive Vit.B12 absorption test, urinary form imino glutamate (FIGLU) and urinary methylmelonate may also provide an important clue in the diagnosis of megaloblastic anemia. Most of these investigations are expensive and require special equipments, materials and expertise which may not be available everywhere.

Gross elevation of serum Lactate dehydrogenase (LDH) in megaloblastic anemia was first reported by Hess and Gehm [2]. Since then number of workers documented the role of serum LDH in anemias. Winston and Warburton [3] had shown a characteristic reversed LDH isoenzyme pattern ie LDH1 > LDH2 in megaloblastic anemia. Besides electrophoresis of serum LDH isoenzyme, number of other methods like heat Inhibition test, acetone inhibition test and chloroform inhibition test are useful to demonstrate the preponderance of fast moving isoenzymes, LDH1 and LDH2.

Being at the interface of a clinical and laboratory discipline the hematologist has to interpret the hematological disorders and to achieve the maximum return as economically and efficiently as possible value for money is a major consideration.

The present study was undertaken to evaluate the significance of total serum LDH in the diagnosis of megaloblastic anemia.

Aim and Objectives:

- To evaluate the utility of serum lactate dehydrogenase in the diagnosis of megaloblastic anemia.
- To study the comparison between serum lactate dehydrogenase levels, peripheral smear and bone marrow examination in macrocytic anemia, where bone marrow is taken as a gold standard.
- To study the correlation between serum lactate dehydrogenase and hemoglobin concentration in megaloblastic anemia.

Materials and methods:

This is a cross sectional study and was carried out in the department of Pathology, MGM medical college and hospital, Aurangabad [MS], India during period from Oct. 2013 to Oct. 2015.

In the present prospective study total 100 cases were study out of which 50 healthy adults were taken as controls. 50 cases of megaloblastic anemia diagnosed on peripheral blood smear and bone marrow examination were taken.

Selection of cases done with patients having Hemoglobin in male <13gm/dl and in female <12gm/dl.

**Inclusion and Exclusion criteria**

**Inclusion criteria:**

- Patients with age more than 13 years are included in study.
- All patients with macrocytic anemia were identified when peripheral blood showed:
  - A mean red blood corpuscular volume >95 fl
  - Anemia with a hemoglobin of
    - <13 g/dl in men.
    - <12 g/dl in women.

**Exclusion criteria:**

- Patients other than megaloblastic anemia viz iron deficiency anemia and hemolytic anemia are excluded.
- Patients with hematological malignancies viz leukemia, lymphomas are also excluded.
- Women in third trimester of pregnancy excluded because of ethical concern about subjecting them to bone marrow aspiration.

**Methods of collection of data.**

Data was collected as per the proforma. Blood sample sent to laboratory were taken to study the hematological parameters like:

- Hemoglobin estimation.
- Peripheral blood smear stained by leishmann stain.
- Bone marrow aspiration smears taken and stained with leishmann stain.

**Biological investigation**

- Total serum LDH

**Results:**

The present study was conducted during Oct. 2013 to Oct. 2015 in the Department of Pathology.

<table>
<thead>
<tr>
<th>Gender</th>
<th>Group I</th>
<th>Group II</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>Male</td>
<td>37</td>
<td>74</td>
</tr>
<tr>
<td>Female</td>
<td>13</td>
<td>26</td>
</tr>
<tr>
<td>Total</td>
<td>50</td>
<td>100</td>
</tr>
</tbody>
</table>

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Out of 50 cases, 37 (74%) cases were males while rest 13 (26%) cases were females. Out of 50 controls, 37 cases are males while rest 13 cases were females.

Table 2: Distribution of Subject According to Age-Group:

<table>
<thead>
<tr>
<th>Gender</th>
<th>Group I</th>
<th></th>
<th>Group II</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>%</td>
<td>No.</td>
<td>%</td>
</tr>
<tr>
<td>&lt;20</td>
<td>08</td>
<td>16</td>
<td>08</td>
</tr>
<tr>
<td>21-30</td>
<td>13</td>
<td>26</td>
<td>13</td>
</tr>
<tr>
<td>31-40</td>
<td>08</td>
<td>16</td>
<td>08</td>
</tr>
<tr>
<td>41-50</td>
<td>08</td>
<td>16</td>
<td>08</td>
</tr>
<tr>
<td>51-60</td>
<td>09</td>
<td>18</td>
<td>09</td>
</tr>
<tr>
<td>60-70</td>
<td>03</td>
<td>06</td>
<td>03</td>
</tr>
<tr>
<td>&gt;70</td>
<td>01</td>
<td>02</td>
<td>01</td>
</tr>
<tr>
<td>Total</td>
<td>50</td>
<td>100</td>
<td>50</td>
</tr>
<tr>
<td>Mean ±SD</td>
<td>38.54±17.23</td>
<td>38.54±17.23</td>
<td></td>
</tr>
</tbody>
</table>

Out of 50 cases, 13 (26%) cases were from age group of 21-30 years. Mean age group of patients was 38.54±17.23 years and ranging from 18 to 80 years.

Table 3: Comparison of Mean MCV in Group I & Group II

<table>
<thead>
<tr>
<th>MCV</th>
<th>Mean</th>
<th>SD</th>
<th>Z-value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I</td>
<td>107.12</td>
<td>10.12</td>
<td>12.80</td>
<td>P=0.000 S</td>
</tr>
<tr>
<td>Group II</td>
<td>87.76</td>
<td>3.45</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

In this study the mean MCV value was 107.12 whereas in controls the mean MCV value was 87.76.

Table 4: Comparison of Mean LDH in Group I & Group II

<table>
<thead>
<tr>
<th>LDH</th>
<th>Mean</th>
<th>SD</th>
<th>Z-value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I</td>
<td>2396.04</td>
<td>1327.23</td>
<td>11.29</td>
<td>P=0.000 S</td>
</tr>
<tr>
<td>Group II</td>
<td>316.3</td>
<td>68.57</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Mean serum LDH calculated in this study was 2396.04 whereas it was 316.3 in control group. The above table depicts that there was marked increase in serum LDH up to 5 times of the upper normal limit of serum LDH which was statically significant with p value (p=0.000).

Table 5: Neutrophils in Group I

<table>
<thead>
<tr>
<th>Neutrophils</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyper segmented</td>
<td>38</td>
<td>76</td>
</tr>
<tr>
<td>Normal</td>
<td>12</td>
<td>24</td>
</tr>
<tr>
<td>Total</td>
<td>50</td>
<td>100</td>
</tr>
</tbody>
</table>

In this study hyper segmented neutrophils were found in 38 (76%) patients. Normal neutrophils seen in 12 (24%) patients.

Table 6: Correlation of LDH & HB in Group I:

<table>
<thead>
<tr>
<th>Correlation between</th>
<th>r-value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>LDH Vs HB</td>
<td>-0.426</td>
<td>P=0.002 S</td>
</tr>
</tbody>
</table>

In this study there was an inverse relationship between serum LDH & Hb which was statistically significant with p value (p=0.002).
Discussion:
Megaloblastic anemia is a significant cause of ill health in developing countries like India. It is suggested that megaloblastic anemia has been highly underestimated in our country and customary practice of treating anemia with iron supplementation with subtherapeutic dosage of folic acid and vit B12 may not provide the required correction of hemoglobin [3]. It is known that marked increase in serum LDH in megaloblastic anemia is due to intramedullary destruction of megaloblasts and high content of LDH in megaloblast. Thus serum LDH can provide an important clue in the diagnosis of megaloblastic anemia.

The present study was undertaken to highlight the serum LDH in diagnosis of megaloblastic anemia diagnosed on peripheral blood smear and bone marrow examination.

Gender distribution
Out of 50 controls which were healthy adults, 26 (74%) were males while 13 (26%) were females, maximum controls were from 21-30 years of age. Out of 50 cases of megaloblastic anemia 37(74%) cases were male, while 13 (26%) were female showing male preponderance. M/F 2:1 ratio.

Labhotia M et al [3] had also shown that 30% cases of megaloblastic anemia were male in comparison to 18.5% which were female with male predominance. Phurailatpam Madhubala Devi et al [4] in study of 50 cases, showed that maximum number of cases were observed in the 21-40 years age group with the male to female ratio 1.5:1 with male predominance. Rajesh para et al [5] in a study of 58 cases of pancytopenia showed 30 males and 28 females with ratio of 1:1.

Age distribution:
In present study maximum numbers of patients were in 21-30 years age group, mean age in megaloblastic anemia was 38.5 ± 17.23 years. Mwando et al [6] showed mean age as 37.7 years in total of 343 patients. Khanduri U et al [7] showed that mean age group was 10-30 years of total 175 patients. Prem kumar M et al [8] in study of 140 patients showed the mean age was 32.8 years with male to female ratio 1.9:1 with male predominance. Ghore B.P et al [9] in retrospective study of serum LDH in megaloblastic anemia showed that mean age group of total 42 patients was 35.12 years with standard deviation ± 17.81.

Incidence & prevention:
In present study it was observed that majority of patients of megaloblastic anemia i.e 78% had predominantly macrocytic anemia and 76% Cases of megaloblastic anemia had hypersegmented neutrophils in peripheral blood smear examination. Labhotia M et al [3] had also shown that majority of patients of megaloblastic anemia had macrocytic anemia and hyper segmented neutrophils were seen as 61%. Uma Khanduri et al [10] showed that 75% patients had hyper segmented neutrophils and majority of patients had macrocytic anemia.

Mean MCV:
In present study, the mean MCV value is 107.12 fl. Parul kodan et al (2014) [11] in his study of clinical profile in patients of pancytopenia at a tertiary care center in South India showed that mean MCV value was 108.44 in total of 44 patients studied. Khanduri U et al (2007) [6] shown mean MCV value was 77-123 fl. Gore et al (2015) [9] showed mean MCV was 115 fl of total 42 patients studied.

Correlation between serum LDH & Hb concentration:
In megaloblastic anemia low value of hemoglobin is associated with disproportionally greater increase in total serum LDH level. In present study mean hemoglobin concentration was 5.25 gm/dl ± 1.53 gm/dl. Gronvell C et al (1961) [12] had also shown that there was an inverse relationship in megaloblastic anemia i.e. low values Hb values are associated with disproportionately greater increase in serum LDH level.

Prem Kumar M et al (2012) [7] in study showed mean Hb level in all patients was 5.3 ± 1.69/dl. Also showed inverse relationship.

Gore et al (2015) [9] mean Hb in these study of 42 patients showed mean Hb as 5.41 ± 1.11. Serum LDH was elevated in 38 patients (90%) and showed inverse relationship between LDH & Hb value.

Serum LDH:
In present study mean serum LDH was 2396.04 IU/L with significant P value (0.000). It means that, it was raised up to 5-8 times the upper normal limit. Hess and Gehn (1955) [2] showed that LDH level raises 5-21 times upper normal limit of serum LDH.

Conclusion:
LDH levels in patients with megaloblastic changes in bone marrow were raised above normal. Hence serum LDH might be used as a screening tool before doing any other diagnostic procedure. Thus we conclude that serum LDH may have an important role in diagnosis of megaloblastic anemia.

References:


Conflict of interest:
None declared.
No source of funding.