Introduction

Pernicious anaemia (PA) is an autoimmune disease in which there is atrophy of gastric mucosa causing destruction of parietal cells and thus decreased secretion of intrinsic factor. This causes vitamin B12 deficiency due to decreased absorption from ileum and results in megaloblastic anaemia. PA is characterized by intrinsic factor antibodies (IFA) and parietal cell antibodies (PCA) which are present in the serum and/or gastric juice of the patient. IFA are specific antibodies which are used to establish the diagnosis of pernicious anaemia. IFA are of two types: type-I IFA inhibits the binding of B12 to intrinsic factor and type-II IFA inhibits binding of intrinsic factor to its ileal binding site. Type-I antibody has been reported in 31 - 76% of patients with pernicious anaemia while type-II antibody is of less diagnostic importance as it occurs less frequently. PA mainly occurs in elderly patients, but younger patients may also be affected. According to a study about 15% patients present in younger age. PA is more common in females than males (1.6:1). Usually patients, give a positive family history. These patients have 2 - 3% increased risk of carcinoma of stomach as compared to patients without pernicious anaemia. Clinically, the patient presents with features seen in megaloblastic anaemia i.e. generalized weakness, jaundice, pallor, lethargy, palpitation, vomiting, diarrhea, numbness and tingling of hands and feet. The most serious consequence of vitamin B12 deficiency is neurological damage which if developed is irreversible, therefore, its early diagnosis, identifying its cause, proper treatment and education of the patient is necessary for its prevention. Pernicious anaemia is an important cause of vitamin B12 deficiency so measurement of IFA is essential for its diagnosis. Patients with pancytopenia and megaloblastic changes need to be investigated for vitamin B12 and/or folate deficiencies, as these are the main causes of megaloblastic anaemia. Vitamin B12 deficiency is the major cause of megaloblastic anaemia in 78% of patients in Pakistan. Therefore, vitamin B12 deficient patients should be investigated for IFA which is specific for the diagnosis of pernicious anaemia.

Methodology

The study was performed at Fauji Foundation Hospital, Rawalpindi, in collaboration with Foundation University Medical College and Armed Forces Institute of Pathology, Rawalpindi, from January 2011 to June 2012. A total of 120 patients of vitamin B12 deficient were
included in this study. Blood sample was collected from each patient for obtaining serum for vitamin B12 level and presence of intrinsic factor antibody.

The vitamin B12 level was measured by chemiluminescence using a commercial kit (Roche Diagnostic GmbH, USA). The analyzer (Cobas 411 elecsys e analyzer) was used for the measurement of vitamin B12 level. The reference range of vitamin B12 was 191 - 663 pg/ml as provided in the literature with reagents. The serum vitamin B12 level below 191 pg/ml was considered as vitamin B12 deficiency.

The test for intrinsic factor antibody was performed by ELISA as per procedure recommended by manufacturer (Anti-intrinsic Factor Antibody ELISA IMMCO diagnostics, USA). The result was reported as positive or negative and expressed in ELISA units per milliliter (EU/ml). The cutoff for positive test was more than 20 EU/ml and for negative test less than 20 EU/ml.

The patients were divided into five groups on the basis of their age for presence of pernicious anaemia. The data was analyzed by using Statistical Package for Social Sciences (SPSS) version 14.

TABLE I: Demographic details of the patients.

<table>
<thead>
<tr>
<th>Pernicious anaemia</th>
<th></th>
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<tbody>
<tr>
<td>Negative</td>
<td>104</td>
</tr>
<tr>
<td>Positive</td>
<td>16</td>
</tr>
</tbody>
</table>

Mean age at presentation (years) range (12-81) range (15-77)

Male to female ratio 30/74 2/14

DISCUSSION

Pernicious anaemia is a common cause of megaloblastic anaemia worldwide especially in population of European and African descent while dietary deficiency of vitamin B12 is common in the Indian subcontinent, Mexico, Central and South America and some areas in Africa. The prevalence of vitamin B12 deficiency in industrialized countries is around 2 - 20% in general population.

The prevalence of PA in general population is 2-3% in European patients. In this study, pernicious anaemia was found in 13.3% (16/120) of vitamin B12 deficient patients as we selected only vitamin B12 deficient patients.

A study from Australia reported a gradually increasing incidence of PA from 2.5% in the third decade to 12% in the eighth decade. In this study, the patients were divided into five groups according to age. A gradually increasing number of patients with increasing age was observed as shown in the Table II. The highest number of patients was seen in the age > 60 years. Another recent study on PA showed that it is a disease of the elderly patients. In their series of 177 patients, 4% patients of PA were below 30 years of age, 10% were between 30 - 40 years and 34% patients were between 60 - 70 years, showing the same increasing frequency with age as seen in this study. The mean age of patients of pernicious anaemia at presentation in this study was 41.5 years which was comparable to another study showing mean age of 45 years.

It is interesting to note that children (age group less than 15 years) also have pernicious anaemia (2/22) in this study. Lahner et al. reported a 15% incidence in young patients. This suggests that pernicious anaemia is not entirely a disease of elderly patients. It may be undiagnosed in younger patients.

PA is more common in females and in first degree relatives of patients. Chan et al. studied 181 Chinese patients of pernicious anaemia and found male to female ratio of 1:1.5. Carmel studied elderly patients and found the prevalence was higher in female (2.7%) as compared to male patients (1.4%). The present results also supports the previous studies, showing male to female ratio of 1:2.5.

Iqbal et al. reported 153 patients of vitamin B12 deficiency including 86 males and 67 females. In contrast, this study showed more of female 88 (73.3%) than male patients 32 (26.6%). This may be due to the reason that the main patients’ bulk in the hospital is found by families of retired army personnel, so relatively more female patients report to study centre hospital.

IFA test was selected as a marker for the detection of pernicious anaemia because it is more specific for the diagnosis of pernicious anaemia than parietal cell antibody (PCA). Previous study showed that specificity of IFA is 40 - 60% for pernicious anaemia which rises to 60 - 80% with the duration of disease. PCA is not...
present as often in pernicious anaemia as it was reported in the past so it does not help in the diagnosis of pernicious anaemia.\textsuperscript{21}

The prevalence of pernicious anaemia has been reported to be 3 - 5 fold higher in autoimmune diseases like type-1 diabetes mellitus and autoimmune thyroid disease.\textsuperscript{22,23} However, no clinical evidence of any autoimmune disorder was found in these patients with pernicious anaemia.

This study supports that pernicious anaemia is not entirely a disease of older age and patients may present in early age. Those presenting with features of megaloblastic anaemia should be investigated for cobalamin and folate deficiency. In case of cobalamin deficiency, further investigation for intrinsic factor antibody to detect pernicious anaemia should be performed so that early treatment is started accordingly to prevent complications.

**CONCLUSION**

The prevalence of pernicious anaemia was 13.3% in vitamin B12 deficient patients. The male to female ratio was 1:2.5. It was relatively more common in patients more than 60 years of age.

**REFERENCES**


