



## Clinical Profile of Vitamin B12 Deficiency and Response to Treatment

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### Abstract

**Background:** Vitamin B12 deficiency is very common in the general population of India. Subclinical B12 deficiency could be easily missed in clinical practice, as the symptoms are vague and numerous and are often overlooked or ignored due to lack of awareness. Early B12 deficiency can be identified by studying the dietary habits of the patients, because the root cause of nutritional deficiencies lies in their poor dietary habits, about which they are not aware of. It will be a good clinical practice to identify the deficiency early and advice on balanced diet which in the long run would bring about a social change.

**Objectives:** To identify the common presentations of B12 deficiency, to make a strong clinical suspicion of B12 deficiency based on the pitfalls in dietary habits, to initiate early treatment, to assess the response to treatment in patients with established B12 deficiency and to prevent recurrence.

**Methods:** This was a prospective study of 203 patients with clinical and hematological evidence of B12 deficiency who attended the outpatient department of PVS Hospital Calicut. The patients were evaluated for their symptoms, dietary practices, physical signs and laboratory tests including Vitamin B12 levels.

**Results:** Fatigue was the most common symptom, seen in 65% of patient's and was apparently due to anemia suggesting that most common clinical presentation can still be hematological. Anorexia was the next common and was seen in 57% of patients, which could have been multifactorial including gastric mucosal atrophy. Skin hyperpigmentation was seen in 40% of patients. Pallor was present in 70% of patients and Romberg's sign was present in 71% of patients. Evidence suggestive of peripheral neuropathy was seen in 25% and psychiatric manifestations including irritability, depression, memory disturbances were seen in 23% of patients. Neuropsychiatric manifestations were seen independent of any hematological manifestations in B12 deficiency. B12 assay showed low levels in 65% of patients only. Majority (67%) of the study group with B12 deficiency had poor intake of fruits and vegetables too and 52% of them had deficient meat or other protein intake.

**Conclusion:** B12 deficiency can present with any of the numerous manifestations affecting hematopoiesis, skin & mucosa and the entire nervous system, including mental functions. Overt B12 deficiency with classical signs and symptoms can be picked up easily, but subclinical cases are easily missed. All patients will not present with classical hematological or neurological manifestations. The key is to suspect the deficiency by remembering the most common symptoms and their correlation with the dietary habits. Since the awareness and practice of a balanced diet is very poor in our society, B12 deficiency can be seen very commonly in clinical practice but is easily overlooked unless we look for it. We miss them because the 'eyes will not see what the mind doesn't know'.

**Keywords:** Vitamin B12; Balanced Diet; Hyperpigmentation; Health education

### Introduction

Vitamin B12- is a water-soluble vitamin, essential for the normal metabolism of the human body. B12 deficiency is very common, the reasons are too many but lack of a balanced diet is the primary reason, but the problem is linked to lifestyle habits, the socioeconomic and cultural issues prevalent in our community [1]. Since almost 98% are not even aware of what constitutes a balanced diet, malnutrition is extremely common in India [1]. Malnutrition cannot be determined by the physique of a person; even obese people are malnourished in terms of proteins and the micronutrients like B12, folic acid and Vitamin D. There are limitations for the laboratory methods available, for diagnosis of nutritional deficiencies including B12 Deficiency. The clinical presentations are numerous, affecting all the organ systems particularly blood, bone marrow, skin, mucosa and the

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entire nervous system including autonomic and peripheral nerves. The symptoms are thus vague, numerous and variable from person to person and are often overlooked due to lack of awareness of the manifestations. Our understanding of this vitamin is not really complete. Vitamin B12 deficiency cannot be diagnosed unless there are clear clinical guidelines or sensitive and specific laboratory methods for the same [2]. Laboratory diagnosis is not always reliable due to numerous reasons [1]. The typical clinical features of B12 deficiency are very rare and appear very late; hence early diagnosis is quite difficult with physical findings and laboratory alone. To complicate the issues further, the underlying diseases causing its deficiency may modify the symptoms of B12 deficiency. Thus, there is a need for a modified approach to arrive at early diagnosis. It is eminently possible, utilizing detailed diet history, clinical picture, and the easily accessible automated hemogram together, for arriving at a clinical judgment [1]. Since the manifestations of deficiency are too many, even changes of hematological parameters may sometimes be absent. But screening of asymptomatic individuals, who are likely to have B12 deficiency can be done with a good diet history [1,3,4]. Some patients with potential risk factors for developing vitamin B12 deficiency can be identified by good clinical evaluation and the past medical and surgical history [1]. Identifying and defining a problem is the starting point for correction of all health-related issues. Since laboratory diagnosis of B12 deficiency cannot be completely relied upon, the clinical picture with proper elicitation of dietary history should be familiar to all treating doctors, so that B12 deficiency can be identified early and correction can be advocated.

The objective of the study was to understand the clinical profile of vitamin B12 deficiency, to evaluate the causes of B12 Deficiency, to correlate deficiency state with dietary habits, and to undertake patient education about the widespread deficiency of vitamin B12 in our population. With the data generated we planned to come out with a guideline for early diagnosis based on the observations.

## Materials and Methods

It was a prospective observational study conducted in the General Medicine outpatient section of the PVS hospital Kozhikode. We could enroll 203 patients for the study (98 males and 105 females) who were worked up before in different places and yet with no diagnosis for refractory anemia or cytopenia not responding to treatment or were referred to us for undiagnosed clinical problems during a period of one year. Patients were recruited for evaluation when they had one or more of the following features, a) strict vegetarians or b) patients taking meat less often (less than one serving per week) c) knuckle hyperpigmentation/dark circle around eyes/other hyperpigmentation, positive Romberg's sign, soreness of tongue and other mucosal areas, anorexia due to gastritis, fatigue, peripheral neuropathy, /ataxia a/ dementia a/ depression/premature graying of hair d) Besides the above features which were clinically suspected to be due to B12 deficiency based on their diet history, another inclusion criteria was hemogram showing MCV>90 with normal RDW, or with low Hb and or cytopenia after excluding other causes for it. Detailed History including proper dietary history was taken for every patient. Patients with diagnosed malabsorption syndromes, those who received blood transfusions within one month prior to presentation, those already on vitamin B12 supplementations, those with history of bleeding from any site and other obvious structural and metabolic diseases were excluded.

All the subjects selected were subjected to estimation of serum

B12 Assay. The clinical features in these patients were recorded. Patients were then counseled regarding a balanced diet containing protein, vegetables, and fruits. Patients fulfilling the inclusion criteria with symptoms, signs and or laboratory evidence of Vitamin B12 deficiency were included and they were given Vitamin B12 injection as intramuscular hydroxocobalamin in doses of 1000 microgram daily for five days and weekly once for five weeks, followed by once-a-month injection. Patients were reassessed for a) improvement in symptoms, b) changes made in dietary habits, c) decrease in MCV and improvement in other parameters in hemogram. They were also given advice regarding balanced diet and were kept under follow up for a minimum period of six months. Patients were reviewed during outpatient visits, only those whose symptoms and signs had improved were taken finally for analysis.

## Results and Discussion

Age of the subjects ranged from 23 to 84 years with a median age of 54 years (n=203). Only those with definite features of B12 deficiency and had improved on parenteral B12 were taken for final analysis. Subjects finally enrolled were of two categories, a) patients with definite B12 deficiency as evidenced by their clinical features and low B12 levels (symptomatic with low B12 levels) b) symptomatic with normal B12 levels but responding to B12 therapy and no other possible cause for their symptoms and signs. The signs of B2 deficiency were more common in the older adults and it was obvious that B12 deficiency can be seen more often in them. Any data on malnutrition can be traced back to the society they belong to and therefore, this data can be extrapolated to the entire society to which our study population belongs to.

### Dietary habits in patients with B12 deficiency

The study had included pure vegetarians and non-vegetarians but taking meat very rarely. There was not much of difference between clinical features between these two groups showing that meat consumption less than two servings per week is inadequate to provide B12. Fruits and vegetable intake were poor in 66% of study subjects and only 9% of study subjects were consuming adequate amounts of fruits and vegetables. There was a statistically significant difference in symptoms or signs between those who had poor intake of vegetables and fruits and those who had good intake of vegetables and fruits. Fruits and vegetables being the only natural source of folic



Figure 1A: Hyperpigmentation of skin and mucosa in patients with B12 deficiency.



**Figure 1B:** Hyperpigmentation of skin in B12 deficiency.

acid, this association suggests that one source of B12 and a source of folic acid should be there in a balanced diet to avoid manifestations of B12 deficiency. This indicates another aspect of B12 deficiency, unless folic acid is also made available through diet the manifestations of B12 deficiency would be more pronounced. Even a source of protein intake was poor in 51% of study subjects and only 15% of them had adequate protein intake in some form or other. Studies in India have reported B12 deficiency in up to 70% to 85% of the study population [5-11]. Even among non-vegetarians, consumption of non-vegetarian proteins was very low. Hence it should be emphasized that B12 deficiency occurs in non-vegetarians also unless they consume B12 containing food items regularly. These data give us insight regarding the gross ignorance about balanced diet in the study population. Majority of the study population were ignorant of the five essential components of each meal and the need for consuming them together (namely carbohydrates, proteins, high fiber vegetables, fruits and water). Majority of the study subjects did not know what to eat, and we feel that this only made them land up as patients. It is another issue that many of them have no access to these items due to lack of social empowerment.

### Symptoms and signs of patients with B12 deficiency

The most common symptoms of B12 deficiency were anorexia and fatigue. Fatigue was present in 65% of patients which was obviously due to anemia. Anorexia was seen in 57% of them which could be due to the gastric mucosal atrophy (atrophic gastritis) in severe B12 deficiency but we did not do anything to document it. Pallor was demonstrable in 71% of the subjects in this study. Not every patient expressed the same degree of anemia for each level of cobalamin deficiency. Severely deficient people had surprisingly mild anemia or even lack of it. The mean Hemoglobin level in our study group was 10.1 g/dl. Anemia is always multifactorial and often due to concomitant iron deficiency as well. Another study from India in patients with megaloblastic anemia showed fatigue in 96% of patients and anorexia in 64% of patients [12]. Pallor was present in 100% of their patients that meant they studied only patients with anemia. Classically B12 deficiency may give rise to megaloblastic anemia, and less commonly pancytopenia with unconjugated hyperbilirubinemia due to dyserythropoiesis. But B12 deficiency can present with any one of the numerous manifestations [1]. It was noted in another study that the degree of megaloblastic anemia of B12 deficiency is inversely

correlated with the presence and severity of neuropsychiatric symptoms at the time of presentation [13]. The underlying reason for this inverse relationship was not well described. Hematological manifestations are due to inadequate vitamin B12 resulting in ineffective DNA synthesis and impaired erythropoiesis. In India folate deficiency is an equally important cause of megaloblastic anemia. Combined B12 and folic acid deficiency is more common than B12 or Folate deficiency alone, and 45% of non-anemic individuals have either subnormal level of B12 alone or combined B12-Folate Deficiency [7]. In general, B12 and folic acid deficiency are suspected with MCV more than 100fl [14]. But we had chosen an MCV>90fl in the inclusion criteria not to miss the subclinical cases also, because many of our patients had coexisting iron deficiency as well. If we use MCV more than 100fl that would be missing those with coexisting iron deficiency. This study was aimed at finding the subclinical B12 deficiency as well, so that early treatment could be initiated. We found that 80% of patients with low B12 level had MCV>90fl, showing statistically significant correlation to deficient state. This observation can be used as an indicator for predicting or strengthening the clinical diagnosis of B12 deficiency, in an individual who has a predisposition to develop B12 deficiency, based on a good dietary history, symptoms and signs [1,15]. Rarely some patients with profound cobalamin deficiency do not even have higher MCV values which could be due to concomitant iron deficiency, thalassemia trait or anemia of chronic disease coexisting [16].

Romberg's sign was positive in 71% subjects in our study group and 64% of those with low serum B12 levels had a positive Romberg's sign. Positive Romberg's sign indicates the involvement of the posterior column in a clinical setting of B12 deficiency. If this subclinical involvement of the spinal cord is not identified and treated promptly, in course of time it can progress to severe disease manifestation. Combined degeneration in the spinal cord may be the most typical manifestation of B12 deficiency, but the typical manifestations of common diseases are rare events, as compared to their atypical manifestations and we should be clinically competent to pick up atypical and early presentations, so that we can initiate an early treatment [17].

We found statistically significant association between anorexia, fatigue and psychiatric symptoms besides other manifestations like pallor. This suggests that unless a high index of suspicion is there and unless it is considered by the first contact doctors (we hardly have any family doctor/GPs now in India!), these patients may go to the wrong specialists and get investigated unnecessarily without any conclusions. In fact, all our patients had gone through this phase before coming to us. We had also found a statistically significant association between poor protein intake with fatigue and skin changes due to B12 deficiency. There was statistically significant association between poor intake of fruits and vegetables and anorexia, fatigue and skin changes. These associations strengthen the relationship between lack of balanced diet and disease manifestations. This is because, if the diet is not balanced, the availability of some of the micronutrients would be less and even if one component is there, it will not work in the absence of others. Hemoglobin synthesis needs folic acid, iron and protein besides B12, and myelin synthesis needs protein, fatty acids and folic acid in addition to B12. If any one of them is missing we may get manifestations of all these deficiencies, several cases of refractory anemia we had seen were refractory only because of ignoring this reality. The metabolic pathways inside our body are complex and there may not always be a one-to-one correlation.



What we understood is to diagnose always based on a strong clinical suspicion, and that too based on the awareness regarding most common ways of presentation, in the background of underlying dietary inadequacies elicited by a good dietary history, which most doctors ignore now.

Skin pigmentation was present in 41% of the study population (Figure 1A, 1B). Skin pigmentation due to vitamin B12 deficiency is an overlooked sign and symptom. This has got a high positive predictive value if the dietary history is suggestive or if there is coexisting hematological or neurological manifestations of B12 deficiency. Association between skin pigmentation and B12 deficiency was studied earlier too [10]. We studied knuckle hyperpigmentation, pigmentation of extremities, oral mucosa hyperpigmentation, after ruling out the most possible causes for the same.

### Neuropsychiatric manifestations in B12 deficiency

We found 23% of patients with B12 deficiency having neuropsychiatric symptoms which improved with B12 therapy and a balanced diet. It was reported earlier that neuropsychiatric manifestations due to cobalamin deficiency occurred commonly in the absence of anemia or macrocytosis [18]. A high index of suspicion of B12 deficiency is needed in such patients presenting with myelopathy, cognitive decline and neuropathy. Good dietary history and skin changes will be an important clue towards diagnosis. We had included memory impairment as a symptom to be evaluated in our sample. It was found that patients labeled as dementia of unknown etiology were found to have clinical improvement in terms of cognitive function after B12 supplementation. Most subjects with Alzheimer's disease were found to have low serum vitamin B12. The study strengthens the role of vitamin B12 in higher mental functions. Psychiatric symptoms in the form of irritability, depression, mood changes and memory disturbances were found in 23% (n=46) of our study subjects with low serum B12. They had any one of the above-mentioned symptoms which improved on treatment. Psychiatric disorders in B12 deficiency can include depression, apathy, irritability, dementia, delirium and hallucinations. Low serum B12 was observed in patients with psychiatric symptoms in previous studies too with majority having no significant hematological or neurological manifestations [19-21]. The possibility of psychiatric disorders being caused by B12 deficiency is often overlooked. Interesting feature to note here is better vitamin B12 levels are thought to be associated with a more favorable outcome in depression [22]. In our study group 25% of patients with normal B12 levels, but had other features of deficiency, had psychiatric symptoms which showed definite clinical improvement with treatment. There has been a report of psychotic symptoms deemed to be resistant to psychotropic treatments that dramatically improved after administration of vitamin B12 [22].

Psychiatric manifestations can occur even before the levels of vitamin B12 are below 175 pg/ml. Studies also mention a 'window period' within which treatment may reverse changes, after which axonal demyelination and other pathological changes may be irreversible [1]. Hence a high index of suspicion must be maintained in patients who present with psychiatric symptoms, especially so when they are refractory to conventional psychotropic or antidepressants.

### Estimation of vitamin B12 levels

Though all the subjects in the study had B12 deficiency based on clinical features, dietary history and improvement of their clinical problem with B12 therapy, we had found out that only 63% of patients

had low B12 levels. Previous studies had proven that normal levels of serum B12 do not exclude symptomatic B12 deficiency. Out of 203 patients, 75 patients had normal B12 levels (37%). This underscores the fact that in clinical practice many cases of B12 deficiency are misdiagnosed, if we use B12 levels as the only the diagnostic criteria and early treatment cannot be initiated in that group of patients. In some patients, interference of serum vitamin B12 assays by IF antibodies have been demonstrated [23]. Our conclusion is that all patients with B12 deficiency do not have low B12 level on laboratory estimation, and normal values will not rule out B12 deficiency in any patient. It has been reported also that B12 assays fail to measure low total vitamin B12 concentrations in some samples because of an unknown artifact [24]. One study reported that 25% of patients with pernicious anemia had false normal values on B12 assay [23]. Bruce HR in 2019 reported a case of overt macrocytic anemia with repeated serum B12 measurement yielding normal results but after three years developing symptoms compatible with severe polyneuropathy and subacute combined degeneration of spinal cord [24]. This case report also proves the superiority of clinical suspicion over laboratory estimation in diagnosing B12 deficiency. David B et al. in a study of performance of serum cobalamin assay for diagnosis of cobalamin deficiency reported that the positive predictive value of a low cobalamin levels with that test was 23% only [25].

Malnutrition is a global issue but particularly relevant in developing countries like India which holds 18% of the world's population. Putting on weight by eating excess calories in any form, but with a grossly unbalanced diet, is not identified as malnutrition and hence there is a great misleading perception regarding data on nutrition if we use weight for height and BMI only to assess the health status. When food insecurity exists in a community, sufficient or even excessive calories may be provided by the limited foods available, but the nutritional quality and diversity of food items in the diet may not support a healthy nutritional status due to inadequate micronutrients. This paradox between poverty and obesity occurs throughout the world, especially so in India. Health is a politically challenging issue because the right to have access to 'a standard of living adequate for health and well-being should be a human right problem' [4]. Doctors are the ambassadors of health and should be interested in the patient as a whole, including the socioeconomic status, diet and lifestyle of the patient.

### Conclusion

Vitamin B12 deficiency is common in the population of northern Kerala and goes undetected many times. MCV more than 90 in an appropriate clinical setting can predict B12 deficiency. Dietary history, skin hyperpigmentation and elevated MCV raises a strong clinical suspicion of B12 deficiency and many undiagnosed symptoms can be attributed to that. It is the dietary history that first suggests the possibility of B12 deficiency, and it is supported by clinical features of B12 deficiency which are often vague, and a higher-than-normal MCV. Lack of a balanced diet is the primary reason for B12 deficiency, since those with poor protein intake and decreased fruits and vegetable intake had positive correlation to symptoms. Anorexia and fatigue were the main symptoms in this study group. B12 deficiency can present with any one of the hematological, neurological, psychological or dermatological manifestations alone. Hematological manifestations can be refractory anemia, leukopenia, thrombocytopenia or pancytopenia. Dermatological manifestation in the form of hyperpigmentation was seen in 41% of patients.

Symptoms suggestive of peripheral neuropathy were found in 25% of the study group. B12 deficiency can show a variety of psychiatric manifestations including depression, irritability, and memory disturbances. All clinical features of B12 deficiency need not be present simultaneously, hematological findings can be minimal or absent sometimes vitamin B12 levels may be normal in some patients with deficiency, and hence we cannot rely on B12 estimation alone. Early initiation of treatment will reverse the progressing neurological worsening in B12 deficient patients.

## Recommendations

There is a need to create awareness on balanced diet among all sections of people. The whole society needs empowerment for consuming a proper balanced diet. Doctors need to work for modifying the dietary habits of the patients in every OPD visit. Prescribing a balanced diet and motivating the patients to follow it lifelong should be included in the practice. This needs large number of primary care doctors.

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