

# M.C.V. should not be the only criteria to order vitamin B12 for anemia under evaluation

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Received 23 April 2012; revised 13 July 2012; accepted 30 July 2012

## ABSTRACT

**Introduction:** A strict vegetarian diet has been associated with increased risk of cobalamin deficiency therefore; one would expect a high prevalence of Cobalamin deficiency in India. Erythrocyte indices have been used in the initial evaluation of anemic patients; high Mean corpuscular volume (MCV) is a traditional criterion for folate and vitamin B12 deficiencies. There is no large study of the prevalence of B12 deficiency among patients with normocytosis or microcytosis. **Method:** We retrospectively analyzed the records of serum vitamin B12 and MCV of both inpatients and outpatients at Santokba Durlabhji Memorial Hospital & Research Institute, Jaipur (Rajasthan) during the period from August 2010-April 2011. The study was aimed at identifying the correlation between vitamin B12 level and MCV; and prevalence of vitamin B12 deficiency in this region. **Result & Conclusion:** Every third person is vitamin B12 deficient in the region. There is no correlation between vitamin B12 levels and MCV in majority of the cases. MCV should not be the only criteria for ordering vitamin B12 for patients with anemia under evaluation.

**Keywords:** Vitamin B12; MCV; Deficiency; Prevalence; Investigation

## 1. INTRODUCTION

Attempts to unravel the mystery of Anemia dates back to almost two centuries. During this period Combe [1], Thomas Addison [2], Austin Flint [3] and William Castle [4] made important contributions in this field. A substantial population of India adheres to a vegetarian diet for cultural and religious reasons. A vegetarian diet is considered to promote health and longevity by protecting against conditions such as cardiovascular disease (CVD) and cancer [5], however this diet may be deficient in some nutrients [6,7]. In particular a strict vegetarian diet has been associated with increased risk of Cobalamin deficiency [7,8]. Malnutrition is common in India [9] and

tropical sprue, gastrointestinal infections, and other nutrient deficiencies frequently result in a malabsorptive state [10,11]. Therefore, one would expect a high prevalence of Cobalamin deficiency in India. Indeed, reports from both India [12,13] and studies of Indians living in other countries [14,15] suggest that nutritional Cobalamin deficiency is common. A recent report shows 46.9% of non-anemic adult subjects having subnormal levels of B<sub>12</sub> or Folate-B<sub>12</sub> deficiency being five times more common than folate [16]. Another study showed 60% of vegetarians and 39% of nonvegetarians were B12 deficient [17]. Classic Cobalamin deficiency is associated with megaloblastic anemia and neurologic symptoms [18]. Measurement of serum Cobalamin is the most commonly used biochemical test for diagnosing Cobalamin deficiency [19,20]. Erythrocyte indices have been used in the initial evaluation of anemic patients; high Mean corpuscular volume (MCV) value is a traditional criterion for folate and vitamin B12 deficiencies [21]. There is no large study of the prevalence of B12 deficiency among patients with normocytosis or microcytosis. Our study was aimed at identifying the correlation between vitamin B12 level and MCV and prevalence of vitamin B12 deficiency in this region.

## 2. MATERIAL AND METHODS

We retrospectively analyzed the records of serum vitamin B12 and MCV of both inpatients and outpatients at Santokba Durlabhji Memorial Hospital & Research Institute, Jaipur (Rajasthan) during the period from August 2010-April 2011. The serum vitamin B12 levels were determined by competitive binding Immunoassay on VITROS 3600 Immunodiagnostic System based on enhanced Chemiluminescence, Ortho Clinical Diagnostics, USA which quantitatively measures B12 concentration in human serum and plasma (EDTA or Heparin) to aid the differential diagnosis of anemia. Biological Reference Interval of vitamin B12 levels was 239 - 931 pg/mL. MCV was measured by Sysmex-1800-xi from EDTA sample (Normal Range 80 - 100 fL). Quality assurance in all tests was maintained as per National standards. We

categorized the vitamin B12 results in four groups ranging from <100, 100 - 239, 240 - 499 and 500 - 1000 pg/mL. The MCV was sorted for each group using MS office Excel 2007 and the mean MCV was established for all the four groups.

### 3. RESULTS

During the Nine month period of study we investigated a total number of 2388 serum vitamin B12 level with corresponding MCV values. The seroprevalence of vitamin B12 deficiency is 36.5%. The results reveal that vitamin B12 deficiency would increase MCV (**Table 1**).

Out of 868 patients who were vitamin B12 deficient 10.36% had high MCV (>100 fL). 13.13% had low MCV (<80 fL) and 76.49% were with normal values of MCV (80 - 100 fL) (**Table 2**).

Out of 1520 patients with normal vitamin B12 level 23.81% had low MCV and 3.48% had high MCV and 72.69% were with normal value of MCV (**Table 3**).

### 4. DISCUSSION

The most notable finding is every third person is Vitamin B12 deficient in this region in concordance with study by Kankonkar *et al.* in 2004 [22]. The population of the Indian subcontinent is >1 billion, most of whom consume a diet low in Cobalamin. Isolated reports suggest that Cobalamin deficiency in India is common; however, this problem has received little attention. The national stra-

**Table 1.** Correlation of vitamin B12 and MCV with distribution of patients.

	Vit. B12 (pg/mL)	% of Patients	Mean MCV (fL)
1	<100 pg/mL	0.8	103.8
2	100 - 239 pg/mL	35.7	87.3
3	240 - 499 pg/mL	39	85.1
4	500 - 1000 pg/mL	24.5	85.8

**Table 2.** MCV distribution in patients with low vitamin B12 levels (n = 868).

	No. of Patients		
Low MCV	Normal MCV	High MCV	
114	664	90	

**Table 3.** MCV distribution in patients with normal vitamin B12 levels (n = 1520).

	No. of Patients		
Low MCV	Normal MCV	High MCV	
362	1105	53	

tegies for improving micronutrient intake do not include Cobalamin [20]. In 1934 Wintrobe published synthesis of Red Cell Measurement for diagnosis and classification of Anemia. This classification was based on derived Red Cell Indices. Erythrocyte Indices have been used in initial evaluation of Anemia patients, High MCV values is traditional criteria for B12 and folate deficiency [23]. Wheeler *et al.* suggested that vitamin B12 should be determined in Anemia patients when MCV > 100 fL [24]. Our study also proves inverse relationship of MCV with vitamin B12 deficiency as P. Vant Sant *et al.* [25], Oosterheris *et al.* [26], Mahmoud A. Fora *et al.* [27] did in their respective studies.

We found several interesting observations during our study in which traditional criteria of vitamin B12 and MCV association was not followed in half of the cases. Among 2388 patients only 6% had macrocytosis, 20% had microcytosis and 74% were normocytic. These findings suggest that concomitant iron or other nutritional deficiencies or hemoglobinopathies [21,28,29] may have been responsible for the normal or even low MCV values in some of our anemic patients; such anemias and hemoglobinopathies have been seen commonly in Southeast Asians [21,30-32]. Moreover, the combination in anemia has been reported in the literature [33-35]. Thompson *et al.* reported that 82% of patients with low B12 levels seen at Bellevue Hospital, New York, had MCV values below 95 fL; they suggested that further evaluation of the suspected B12 deficiency should not be deterred by a normal MCV value which is one of the important observation in our study [36].

One study from Vancouver AIDS conference (1996) suggested that MCV does not always get high even if vitamin B12 is low so a normal value does not necessarily means that B12 levels is normal, which correlates with our findings. Thus physicians should not consider elevated MCV as diagnostic criteria for Vitamin B12 deficiency [17,26,37,38]. Oosterhuis *et al.* analyzed the diagnostic value of an elevated MCV for B12 deficiency where the sensitivity was only 17% - 30%, and up to 84% of the deficiency would be missed [26]. Any screening criteria selected would miss a significant number of B12 deficient patients, so there may be a case for universal B12 screening [39].

### 5. LIMITATIONS

Our study has some important limitations. First, dietary information was not available. Second, our study was based on serum B12 results, which alone, without other biochemical markers such as Homocysteine, Methylmalonic acid might not be an accurate measures of B12 deficiency [20]. Even if our findings are valid, we must consider their clinical significance. We believe that B12

deficiency is not just a laboratory finding but a clinically relevant issue as the symptoms are vague and nonspecific but respond to appropriate Vitamin B12 therapy.

## 6. CONCLUSION

Every third person is vitamin B12 deficient in the region. There is no correlation between vitamin B12 levels and MCV in majority of the cases. MCV should not be the only criteria for ordering vitamin B12 for patients with anemia under evaluation.

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