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# Risk Factors and Frequency of Vitamin B12 Deficiency among Patients Visiting Primary Care Clinics in Karachi, Pakistan

Fakhrunnisa Waheed, Mohammad Minhaj, Nida Ghouri, Faridah Amir Ali, Muhammad Junaid Patel

# **ABSTRACT**

**Objective:** To examine the associations of sociodemographic factors, comorbidities, diet, medications, vitamin D levels, hematological indices, and symptoms with vitamin B12 status among primary care patients in Karachi, Pakistan.

**Methods:** This cross-sectional study was conducted in 2023 at the Family and Internal Medicine clinics of Indus Hospital and Health Network. Adults recently tested for vitamin B12 as advised by physicians were enrolled (n=252). Data on demographics, comorbidity, medications, diet, and symptoms were collected through structured interviews, alongside BMI and laboratory results. Associations were analyzed using descriptive statistics, Chi-square, and correlation coefficient, with p<0.05 considered significant.

**Results:** Of 252 participants (71.4% female, median age 46 years), fatigue (79.5%), muscle weakness (73.2%), and numbness/tingling (69%) were the most frequent symptoms in those with B12 deficiency or insufficiency. Decreased appetite (p=0.003) and weight loss (p=0.010) were significantly associated with low B12 levels. Dietary patterns showed strong associations: reduced milk intake (72% deficient) and infrequent red meat consumption (49.6% deficient) were linked to deficiency. Vitamin D correlated positively with B12 levels (r=0.215, p=0.006), while mean corpuscular volume correlated negatively (r=-0.151, p=0.019). No significant associations were found with BMI (p=0.175), comorbidities (p>0.05), or metformin/PPI use (p>0.05).

**Conclusion:** Low vitamin B12 levels were associated with specific dietary insufficiencies, constitutional and neurological symptoms, and hematological indices. These findings highlight the need for heightened clinical awareness and integration of dietary counseling within primary care. These findings highlight the need for targeted screening and interventions to prevent deficiency in underprivileged populations.

**Keywords:** Cobalamin deficiency, B12 deficiency, macrocytic anemia, megaloblastic anemia, nutritional anemia, vitamin B12 deficiency, Pakistan

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#### INTRODUCTION:

Vitamin B12 (Cobalamin) is an essential micronutrient crucial for various physiological processes, hence its deficiency leads to a wide array of clinical manifestations, including hematological, dermatological and gastrointestinal abnormalities, neurological impairment and cognitive decline. The prevalence of vitamin B12 deficiency is common and is more frequent in low- and middle-income countries with a poor or inadequate diet of animal foods. Risk factors for its deficiency have been identified world over, such as old age, vegetarian diet, inflammatory bowel disease, hypothyroidism, history of gastrointestinal resection and prolonged use of medications like proton pump inhibitors (PPI) and metformin. <sup>3,4</sup>

In impoverished countries, insufficient dietary intake is a significant cause of vitamin B12 deficiency.<sup>5,6</sup> For example, certain groups with reduced meat intake due to cultural or societal factors have up to 86.5% vitamin B12 deficiency.<sup>7</sup> Yet, due to variable and vague signs and symptoms, and a high cost of screening, it is often unnoticed by clinicians.<sup>7-</sup>

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Routine screening is not cost-effective, hence the prevalence of vitamin B12 deficiency among healthy individuals in Pakistan remains unknown. A large study conducted among pregnant women found more than half of them to be vitamin B12 deficient, with an association with gestational diabetes mellitus (GDM).<sup>10</sup> Another study showed that 41.9% of adolescents in a vulnerable group had vitamin B12 deficiency.9 A study conducted in Karachi on diabetics revealed vitamin B12 deficiency in 46.3% of patients, out of which 18.4% were on metformin. This was supported by another study that showed a 31% prevalence among diabetics. 11,12 However, another study on another population in Pakistan did not find correlation between vitamin B12 insufficiency and diabetes.<sup>13</sup> Other studies found B12 deficiency to be associated with old age, 14 depression, 15 and dementia, with significant improvement in symptoms following adequate vitamin B12 replacement. 16 Despite the deficiency is common in the Pakistani population, no consistent correlation has been found among the risk factors examined. Also, local studies have reported specific risk factors among various vulnerable populations, multiple risk factors have not been assessed in a single population. This study therefore aims to determine the socio-demographic factors, co-morbid illnesses, medications, symptoms, weight, dietary factors, various blood parameters and vitamin D levels with vitamin B12 insufficiency and deficiency among an underprivileged population presenting to primary care clinics at Indus Hospital and Health Network (IHHN) in Karachi, Pakistan. The findings of this study could contribute towards creating a screening model to detect early B12 deficiency and therefore lead to targeted interventions to help reduce the burden of B12 deficiency and hence associated morbidity in our population.

# **METHODOLOGY:**

This is a cross-sectional study conducted for a year (2023), in Family Medicine and Internal Medicine primary care clinics, at Indus Hospital and Health Network (IHHN), Korangi campus Karachi. IHHN is a free tertiary care health network, catering to the underprivileged population throughout the city. Adult patients who had serum vitamin B12 levels checked within the past one week as advised by the physician, were approached for consent to participate. Patients with malabsorption syndrome, alcoholism, liver disease, malignancies, inflammatory bowel disease, history of gastric/intestinal resection, or those who were unable to give consent (mentally incapacitated), pregnant females, and bed-bound patients were excluded.

The sample size for this study was calculated to be 252 through WHO sample size calculator, using the prevalence of vitamin B12 deficiency in adults 8.78%, <sup>17</sup> margin of error 3.5% and confidence level 95%.

A structured questionnaire was administered to collect data. The data was collected by postgraduate medical trainees of Family Medicine who were trained by the principal investigator. This was completed as a one-on-one interview, in an appropriate private setting. The questionnaire had 7 sections: sociodemographic information, self-reported comorbid illnesses, medication history, eating habits, body mass index (BMI) and available lab parameters. The BMI (kg/m²) was calculated following the WHO standard guidelines, categorized as underweight (<18.5), normal weight (18.5-24.9) and overweight (=25.0).

Risk factors were compared among three groups based on vitamin B12 levels as follows: Normal level: = 400 pg/ml, vitamin B12 insufficiency (200-399 pg/ml) and deficiency (< 200 pg/ml). 3,18

Statistical Analysis: Collected data were analyzed on SPSS version 26, a p-value of less than 0.05 was considered significant. Means (SD) or median (IQR) is reported for quantitative variables such as age, lab parameters and anthropometric measure measurements on the basis of normality. Normality was checked through Shapiro wilk test. Frequency and percentage were reported for categorical variables such as gender, co-morbid illnesses, sociodemographic variables, symptoms, dietary factors and vitamin B12 deficiency. The Mann-Whitney U test was used to analyze differences in continuous variables between independent groups. For categorical data, Chi-square test, with the Fisher-exact test was applied in instances of small sample sizes. After univariate analysis, Pearson correlation was applied to determine a linear relationship between B12 and other quantitative variables such as blood parameters and vitamin D.

Ethical Considerations: For conducting this research, we adhered to the highest ethical standards to ensure the integrity and well-being of all participants. We obtained verbal informed consent from each participant, with a clear explanation of the study's purpose, procedures, and potential risks. This study was approved by Institutional Review Board (IRB) of Indus Hospital and Health Network (IRB Number: IHNN\_IRB\_2022\_06\_005).

# **RESULTS:**

The study included 252 participants with 71.4% females and 28.6% males. The median age of the participants was 46 years (IQR: 35-56). Majority of females were housewives (51%) and a quarter of males were retired. A quarter of the participants were illiterate while almost 50% had not completed 10<sup>th</sup> grade. There was no association of age groups (p=0.170), gender (p=0.310), educational status (p=0.63), and working status (p=0.100) with vitamin B12 deficiency or insufficiency.

Around one-third of participants were had a normal BMI, one third were overweight, while 19.4% were obese and 13.5% were under weight. There was no association of BMI with vitamin B12 levels (p=0.175). (Figure 1)

Among all participants, almost half (48%) were deficient in vitamin B12, 43.7% had insufficiency, while only 8.3% had a normal level. More than half (58.7%) had co-morbid illnesses. Among these, 59.3% had hypertension, 43.4% had diabetes, 19.3% had hypothyroidism, and 7.6% had depression/anxiety. There was no significant difference among distribution of co-morbid illnesses among participants with normal, insufficient or deficient vitamin B12 levels (Table 1) however, hypertension was the more common among vitamin B12 insufficient as compared to deficient patients but p value was not statistically significant (p 0.617). Cardiovascular diseases were most common (44.4%) in the study population followed by hypertension (34.1) and diabetes (25%).

Most common symptoms among patients with vitamin B12 insufficiency and deficiency included fatigue (79.5%), weakness in muscles (73.2%), tingling/numbness (69%), irritability (64.4%), and balance issues in walking (57.7%). Decreased appetite (p 0.003) and weight loss (p 0.010) were more common among participants with B12 insufficiency or deficiency than those with normal B12 levels.

There was no significant association found between vitamin B12 levels and the use of medications, smoking status, or other addictive behaviors, as indicated in Table-2. A trend was observed among participants taking multivitamins, with a higher proportion having normal vitamin B12 levels

Table 1: Association of demographic characteristics with vitamin B12 levels among patients

	B12 Normal N=21	B12 Insufficient N=110	B12 Deficient N=121	Total N=252	P-value	
Age in year						
Median (IQR)	60(31-70)	47(35-57)	44(35-55)	46(35-56)	0.170‡	
Min-Max	17-80	15-80	15-81	15-81	0.179 <sup>†</sup>	
Members in household	1					
Median (IQR)	6(5-10)	6(5-8)	5(4-7)	6(5-6)	0.100	
Min-max	2-20	2-17	1-29	1-29	0.108 <sup>¶</sup>	
Numbers of children						
Median (IQR)	5(2-6)	3(1-5)	3(1-4)	4(1-6)	0.069 <sup>¶</sup>	
Min-max	0-10	0-10	0-10	0-13	0.069	
Gender						
Male	7(33.3)	26(23.6)	39(32.2)	72(28.6)		
Female	14(66.7)	84(76.4)	82(67.8)	180(71.4)	$0.310^{\dagger}$	
Total	21(100)	110(100)	121(100)	252(100)		
Working Status			_			
Employed	3(14.3)	2(1.8)	6(5)	11(4.4)		
Housewife	11(52.4)	62(56.4)	56(46.3)	129(51.2)		
Retired	3(14.3)	30(27.3)	33(27.3)	66(26.2)	$0.109^{\dagger}$	
Unemployed	4(19)	16(14.5)	26(21.5)	46(18.3)		
Total	21(100)	110(100)	121(100)	252(100)		
Education						
Above Matric	7(33.3)	28(25.5)	25(20.7)	60(23.8)		
Matric	7(33.3)	28(25.5)	37(30.6)	72(28.6)		
Under Matric	2(9.5)	29(26.4)	30(24.8)	61(24.2)	0.639 <sup>†</sup>	
Uneducated	5(23.8)	25(22.7)	29(24)	59(23.4)		
Total	21(100)	110(100)	121(100)	252(100)		
BMI status						
Underweight	3(14.3)	13(11.8)	18(14.9)	34(13.5)		
Normal	10(47.6)	30(27.3)	45(37.2)	65(33.7)		
Overweight	6(28.6)	46(41.8)	32(26.4)	83(33.3)	0.175†	
Obese	2(9.5)	21(19.1)	26(21.5)	49(19.4)		
Total	21(100)	110(100)	121(100)	252(100)		

<sup>\*</sup>P-value<0.05, ¶ Mann Whitney U test, † Chi-square test, † Fisher-exact test

<sup>\*</sup>Vitamin B12 Normal level: > 400 pg/ml, insufficiency (200-399 pg/ml) and deficiency (< 200 pg/ml)

		B12 Normal N=21	B12 Insufficient N=110	B12 Deficient N=121	Total N=252	P-value	
Cardiovascular Disease	No	10(47.6)	59(53.6)	71(58.7)	140(55.6)	0.5551	
Carulovasculai Discase	Yes	11(52.4)	51(46.4)	50(41.3)	112(44.4)	0.5551	
Diabetes	No	13(61.9)	79(71.8)	97(80.2)	189(75)	0.101	
Dianetes	Yes	8(38.1)	31(28.2)	24(19.8)	63(25)	0.12 <del>1</del>	
Hypertension	No	15(71.4)	69(62.7)	82(67.8)	166(65.9)	0.6151	
riypertension	Yes	6(28.6)	41(37.3)	39(32.2)	86(34.1)	0.617±	
Hypothyroidism	No	19(90.5)	100(90.9)	105(86.8)	224(88.9)	0.501	
	Yes	2(9.5)	10(9.1)	16(13.2)	28(11.1)	0.591	
Stroke History	No	21(100)	108(98.2)	119(98.3)	248(98.4)	4.	
	Yes	0(0)	2(1.8)	2(1.7)	4(1.6)	1†	
Depression/Anxiety	No	20(95.2)	103(93.6)	118(97.5)	241(95.6)	0.2651	
	Yes	1(4.8)	7(6.4)	3(2.5)	11(4.4)	0.267†	
Coronary Artery Disease	No	19(90.5)	109(99.1)	116(95.9)	244(96.8)	0.0661	
	Yes	2(9.5)	1(0.9)	5(4.1)	8(3.2)	0.066†	
Others	No	14(66.7)	102(92.7)	109(90.1)	225(89.3)	0.002*1	
Onicis	Yes	7(33.3)	8(7.3)	12(9.9)	27(10.7)	0.002*1	

Table 2: Association of comorbidities with vitamin B12 levels among patients

compared to those with B12 insufficiency and deficiency (47.6%, 29.1%, and 25.6% respectively, P=0.086) (Table-2). Results also indicated a significant association between dietary habits and vitamin B12 levels, with a higher proportion of normal B12 patients among those consuming multivitamins, red meat and egg (2-3 times per week). Majority of vitamin B12 deficient patients were those who do not drink milk (Table -3).

The correlation between age and various blood parameters, with B12 levels was computed. Age showed a weak positive correlation with B12 levels (r = 0.091), indicating a slight tendency for B12 levels to increase with age, though statistically not significant (p = 0.148). Conversely, vitamin D exhibited a moderate positive correlation with B12 levels (r = 0.215) (p = 0.006) (Figure 2). Hemoglobin showed a weak positive correlation with B12 levels (r = 0.100) but statistically insignificant (p = 0.121). Mean corpuscular volume (MCV) levels had a statistically significant negative correlation with B12 levels (r = -0.151) (p = 0.019). However, mean corpuscular hemoglobin concentration (MCHC) exhibited no correlation with B12 levels (r = 0.007) (p =0.917). Overall, while some blood parameters showed significant associations with B12 levels, others exhibited weak or negligible correlations, suggesting varied relationships between B12 levels and various blood parameters.

# DISCUSSION:

This study reports an alarmingly high frequency of vitamin B12 deficiency and insufficiency among patients whose B12 levels were requested by the physicians. B12 levels were below normal among 91.6 % of the patients; while

48% had documented deficiency of the micronutrient. Low B12 levels have also been reported in various other studies conducted in specific groups in Pakistan. <sup>9,11,12</sup> The exceptionally high frequency of B12 deficiency in our study is likely, because only patients who were requested by physicians to have their B12 levels checked due to high suspicion of deficiency, were included.

Vitamin B12 plays an important role in various bodily functions; therefore, its deficiency is marked by several symptoms. Lack of energy, weakness in muscles, irritability, numbness/tingling and trouble walking were the most reported patient complaints. These symptoms match the findings reported by Wolfenbüttel and colleagues, who also found improvement in these symptoms post cobalamin therapy for 6 months. Weight loss (56%) was also a significant among the insufficient and deficient group, and it is one of the known gastrointestinal manifestations of low B12 levels. Hence, it is worth reiterating the importance of recognizing these symptoms earlier in at-risk groups.

Our study reports an interesting positive relationship between vitamin D and vitamin B12 levels. As majority of the participants were females, preexisting low vitamin D levels due to less consumption of dairy products and decreased sun exposure in females in our population could provide an explanation for this result. <sup>6,19</sup> This relates to the findings of Karabayir et.al which also conclude that vitamin D deficiency could be common in vitamin B12 deficient patients. <sup>20</sup> More comprehensive studies are needed to reveal the relationship between vit B12 and vit D deficiency.

Vitamin B12 deficiency is a known cause of macrocytic anemia,<sup>3</sup> and our study supported this association. MCV

Table 3: Association of symptoms with vitamin B12 levels among patients

		B12 Normal N=21	B12 Insufficient N=110	B12 Deficient N=121	Total N=252	P-value	
	Yes	12(60)	80(73.4)	83(68.6)	175(70)		
Muscle Weakness	No	7(35)	25(22.9)	36(29.8)	68(27.2)	0.397†	
	Sometimes	1(5)	4(3.7)	2(1.7)	7(2.8)		
	Yes	14(70)	53(48.2)	42(34.7)	109(43.4)		
Dyspepsia	No	4(20)	46(41.8)	68(56.2)	118(47)	0.018*1	
July 1	Sometimes	2(10)	11(10)	11(9.1)	24(9.6)		
	Yes	14(70)	46(41.8)	52(43)	112(44.6)		
Numb or tingling feeling in hands	No	4(20)	57(51.8)	63(52.1)	124(49.4)	0.102t	
	Sometimes	2(10)	7(6.4)	6(5)	15(6)		
	Yes	15(75)	51(46.4)	72(59.5)	138(55)		
Trouble walking	No	5(25)	51(46.4)	49(40.5)	105(41.8)	0.003*†	
C	Sometimes	0(0)	8(7.3)	0(0)	8(3.2)	·	
	Yes	7(35)	24(21.8)	27(22.3)	58(23.1)		
Nausea	No	11(55)	75(68.2)	87(71.9)	173(68.9)	0.425†	
	Sometimes	2(10)	11(10)	7(5.8)	20(8)		
	Yes	14(70)	32(29.4)	48(39.7)	94(37.6)	0.003*†	
Decreased appetite	No	5(25)	75(68.8)	70(57.9)	150(60)		
**	Sometimes	1(5)	2(1.8)	3(2.5)	6(2.4)		
	Yes	7(35)	40(36.7)	64(53.8)	111(44.8)		
Weight loss	No	13(65)	69(63.3)	53(44.5)	135(54.4)	0.022*†	
_	Sometimes	0(0)	0(0)	2(1.7)	2(0.8)		
	Yes	15(75)	65(59.6)	74(61.7)	154(61.8)	0.317±	
Irritability	No	4(20)	36(33)	43(35.8)	83(33.3)		
	Sometimes	1(5)	8(7.3)	3(2.5)	12(4.8)		
	Yes	19(95)	85(78)	86(71.7)	190(76.3)		
Fatigue	No	1(5)	23(21.1)	33(27.5)	57(22.9)	0.14†	
	Sometimes	0(0)	1(0.9)	1(0.8)	2(0.8)		
	Yes	10(50)	20(18.3)	18(15)	48(19.3)		
Diarrhea	No	9(45)	76(69.7)	93(77.5)	178(71.5)	0.01*†	
	Sometimes	1(5)	13(11.9)	9(7.5)	23(9.2)		
	Yes	6(28.6)	8(7.3)	9(7.6)	23(9.2)		
Smooth and tender tongue	No	15(71.4)	99(90.8)	108(90.8)	222(89.2)	0.06†	
	Sometimes	0(0)	2(1.8)	2(1.7)	4(1.6)		
Fast heart rate	Yes	8(38.1)	46(42.2)	56(46.7)	110(44)		
	No	13(61.9)	46(42.2)	52(43.3)	111(44.4)	0.182 <del>1</del>	
	Sometimes	0(0)	17(15.6)	12(10)	29(11.6)		
	Yes	3(14.3)	10(9.1)	9(7.5)	22(8.8)		
Reduced Smell	No	17(81)	97(88.2)	110(91.7)	224(89.2)	0.346†	
	Sometimes	1(4.8)	3(2.7)	1(0.8)	5(2)		
	Yes	12(57.1)	36(33)	31(25.8)	79(31.6)		
Forgetfulness	No	7(33.3)	58(53.2)	83(69.2)	148(59.2)	0.004*1	
	Sometimes	2(9.5)	15(13.8)	6(5)	23(9.2)		

<sup>\*</sup>P-value<0.05, ¶ Mann Whitney U test, † Chi-square test, † Fisher-exact test

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Table 4: Association of medications and smoking with vitamin B12 levels among patients	ons and smoking with vitamin B12 levels among patients
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	B12 Normal N=21	B12 Insufficiency N=110	B12 Deficiency N=121	Total N=252	P-value			
Use of Protor	Use of Proton Pump Inhibitors							
Yes	3(15)	12(11.1)	6(5.2)	21(8.6)				
No	17(85)	96(88.9)	109(94.8)	222(91.4)	$0.168^{\dagger}$			
Total	20(100)	108(100)	115(100)	243(100)	1			
Use of Metfo	rmin							
Yes	5(62.5)	19(61.3)	14(58.3)	38(60.3)				
No	3(37.5)	12(38.7)	10(41.7)	25(39.7)	$1.000^{\dagger}$			
Total	8(100)	31(100)	24(100)	63(100)	1			
Currently on	Pregabalin/ D	uloxetine			•			
Yes	-	5(4.7)	1(0.9)	6(2.5)				
No	19(100)	102(95.3)	113(99.1)	234(97.5)	0.493 <sup>†</sup>			
Total	19(100)	107(100)	114(100)	240(100)				
Smoke Cigar	ettes (at least o	nce a day for a mont	h in the last 1 year	r)				
Yes	1(4.8)	2(1.9)	4(3.3)	7(2.8)				
No	20(95.2)	106(98.1)	116(96.7)	242(97.2)	0.279 <sup>†</sup>			
Total	21(100)	108(100)	120(100)	249(100)				
Use of Tobac	Use of Tobacco on regular basis							
Yes	-	13(11.8)	14(11.6)	27(10.7)				
No	21(100)	97(88.2)	107(88.4)	225(89.3)	$0.525^{\dagger}$			
Total	21(100)	110(100)	121(100)	252(100)	1			
Use Beetle nut								
Yes	3(14.3)	17(15.5)	20(16.5)	40(15.9)				
No	17(81)	91(82.7)	89(73.6)	197(78.2)	0.109 <sup>†</sup>			
Sometimes	1(4.8)	2(1.8)	12(9.9)	15(6)	0.109			
Total	21(100)	110(100)	121(100)	252(100)	1			
	1	<u> </u>						

increased as B12 levels decreased, indicating a negative correlation between the two parameters.

Adequate vitamin B12 can be obtained through diets rich in this micronutrient, such as red meat and milk. Therefore, certain groups who avoid these sources due to cultural, religious, or financial reasons are more at risk of developing B12 deficiency. We found that 72% of those who do not drink milk, and 49.6% of those who consumed red meat less than once per week or consumed no red meat at all had documented vitamin B12 deficiency. Our findings are in line with the report of Bacha and colleagues, 6 who reported reduced diversity in food sources and reduced milk consumption in the Pakistani population.

There was no correlation of BMI or co-morbid illnesses with vitamin B12 levels, however the frequency of patients with known cardiovascular diseases, hypertension and diabetes among our study population was 44%, 34% and 25% respectively. These numbers are concerning especially in a population with meager resources and heath care access, and warrants a need for nationwide awareness and interventions to screen and prevent non-communicable diseases.

Further there was no correlation of B12 deficiency with medication usage (metformin and proton pump inhibitors) (PPI) in our results. This is surprising as vitamin B12 deficiency is a well-known side effect of these medications. A possible explanation could be that the duration and dosage of metformin or PPI use were not systematically recorded in our study, and deficiency risk increases with long-term or high-dose exposure. Moreover, B12 deficiency is multifactorial and may be influenced by dietary patterns, malabsorption, age, or co-morbid conditions, which may have masked any direct association with these medications in our sample. Additionally, the relatively small number of participants with normal vitamin B12 levels could have further diluted the association of variables.

# **CONCLUSION:**

This study underscores the clinical importance of recognizing vitamin B12 deficiency in underprivileged primary care populations. Dietary insufficiency and nonspecific symptoms remain key challenges, while laboratory correlations highlight the complexity of its detection. Although limited by selection bias and lack of generalizability, the findings emphasize the need for greater clinical awareness, integration of dietary

Table 5: Association between the use of multivitamins and dietary intake with vitamin B12 levels among patients

	B12 Normal N=21	B12 Insufficiency N=110	B12 Deficiency N=121	Total N=252	P-value		
Use of Multivitamins (for last 3 months)							
Yes	10(47.6)	32(29.1)	31(25.6)	73(29)			
No	11(52.4)	78(70.9)	90(74.4)	179(71)	$0.086^{\dagger}$		
Total	21(100)	110(100)	121(100)	252(100)	0.000		
Use of Red Meat							
Not at all	8(38.1)	23(20.9)	21(17.4)	52(20.6)			
Less than once a week	3(14.3)	36(32.7)	39(32.2)	78(31)	1		
At least once a week	2(9.5)	24(21.8)	35(28.9)	61(24.2)	$0.072^{\dagger}$		
2-3 times per week	8(38.1)	23(20.9)	25(20.7)	56(22.2)	1		
Almost daily	-	4(3.6)	1(0.8)	5(2)	1		
Total	21(100)	110(100)	121(100)	252(100)	1		
Use of Egg							
Not at all	6(28.6)	40(36.4)	38(31.4)	84(33.3)			
Less than once a week	3(14.3)	27(24.5)	26(21.5)	56(22.2)	]		
At least once a week	2(9.5)	18(16.4)	26(21.5)	46(18.3)	0.338†		
2-3 times per week	8(38.1)	16(14.5)	20(16.5)	44(17.5)	0.556		
Almost daily	2(9.5)	9(8.2)	11(9.1)	22(8.7)	]		
Total	21(100)	110(100)	121(100)	252(100)			
Use of Milk on a daily basis							
Not at all	12(57.1)	63(57.8)	87(72.5)	162(64.8)			
Less than once a week	2(9.5)	9(8.3)	8(6.7)	19(7.6)			
At least once a week	2(9.5)	13(11.9)	8(6.7)	23(9.2)	0.227†		
2-3 times per week	-	10(9.2)	7(5.8)	17(6.8)	] ".22"		
Almost daily	5(23.8)	14(12.8)	10(8.3)	29(11.6)	]		
Total	21(100)	109(100)	120(100)	250(100)			

Table 6: Correlation of Vitamin B12 with Vitamin D

	R-Value	p-value
Vitamin D	0.215	0.006

counseling, and development of cost-effective screening strategies in primary care to enable earlier diagnosis and management

## LIMITATION:

Our study was limited to patients presenting to the primary care clinics of a single charitable hospital, which restricted recruitment primarily to individuals from low or lower middle income groups, thereby reducing the generalizability of the findings. In addition, the inadequate number of participants with normal vitamin B12 levels (control group) prevented us from conducting a case-control study, which would have been a more suitable design. Lastly, due to limited funds for testing B12 levels according to study protocol, we were only able to include patients who had already been advised by their physicians to undergo B12 level assessment, hence the actual prevalence could not be

calculated and the frequency of abnormal B12 levels is exaggerated.

#### **DECLARATION:**

The datasets generated and/or analysed during the current study are not publicly available but are available from the corresponding author on reasonable request

# | Authors Contribution:

**Fakhrunnisa Waheed:** Conceptualization, study design, and supervision.

Mohammad Minhaj: Data collection and analysis.

**Nida Ghouri:** Literature review and methodology.

Faridah Amir Ali: Manuscript writing and editing. Muhammad Junaid Patel: Data interpretation and final

manuscript review

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