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Original Research

Evidence from a tertiary hospital in Jharkhand, "Unravelling the Interplay of Diet and Comorbidities in Vitamin B12 Deficiency: Implications for Haematological Health in a Diabetic Cohort

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Abstract

Introduction - Vitamin B12 deficiency is a common comorbidity among people with diabetes, and the way it interacts with diet and other health conditions can impact blood health. This study set out to explore these connections in a group of individuals with type 2 diabetes.

Methods We included 420 diabetic patients in Jharkhand, India, who were found to have low levels of vitamin B12. We gathered information regarding their diets, any existing anemia, harmful habits, medical histories, B12 levels, and blood-related metrics. We then used appropriate statistical tests to analyze the data. JAMOVI latest version was used to analyse the data. $p<.05$ was considered statistically significant .

Results Our findings indicated that those with pre-existing anemia and other deficiencies were more likely to experience severe vitamin B12 deficiency. We also discovered that severe deficiency was linked to increased mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH). Meanwhile, having anemia was associated with lower levels of hemoglobin, red blood cell count, lymphocytes, monocytes, and MCV, but higher levels of neutrophils and MCHC. Interestingly, we didn't find significant links between a person's dietary habits or malabsorption symptoms and how severe their B12 deficiency was.

Conclusion - The study indicates that while managing B12 deficiency in diabetic patients, knowledge of current anaemia and other deficiencies is absolutely vital since these elements might affect the degree and consequence of the deficiencies on distinct hematological profiles. This emphasises the need of taking into account dietary habits and medical conditions in effective management of these patients.

Introduction

A chronic metabolic condition affecting millions all over the world, diabetes mellitus is characterised by hyperglycemia resulting from abnormalities in either insulin production, insulin action, or both (1). Diabetes' global prevalence has risen dramatically; estimates from the Global Burden of Disease (GBD) 2021 indicate a significant impact on mortality and morbidity across the globe (2). Given a large and growing diabetic population, which creates significant challenges for the healthcare system , the situation in India is particularly concerning (3). Apart from the well-known macro- and microvascular issues, those with diabetes are also susceptible to certain micronutrient deficiencies, including vitamin B12 deficiency (4).

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A vital vitamin for haematopoiesis, neurological function, and DNA synthesis is vitamin B12, which is also known as cobalamin (5). The main source of this vitamin is dietary consumption, mostly from animal sources. But certain individuals might not consume enough because of dietary habits like vegetarian diets common in many areas of India (6). Furthermore, metformin, a frequently recommended antidiabetic drug, has been linked to poor B12 absorption in diabetic patients (7). The interaction between dietary patterns and drugs used in the framework of diabetes could consequently increase the risk of vitamin B12 insufficiency.

Known worldwide, vitamin B12 deficiency is a public health concern especially for disadvantaged groups since it causes anaemia and neurological diseases. T hehaematological effects of B12 deficiency range from macrocytic anaemia to more subtle alterations in red blood cell indices and perhaps influence other blood cell lineages (9). In diabetic individuals, the existence of comorbidities such pre-existing anaemia, poor lifestyle choices, and other underlying medical issues could further complicate the link between B12 status and haematological health. Mercantepe's research also drew attention to the intricate interaction between B12 levels, obesity, and diabetes (10).

The combination of a sizable diabetes population and prevalent vegetarian diet habits in India provides a particular background for research on vitamin B12 insufficiency. Moreover, the frequency and effects of this deficiency could be affected by regional differences in dietary practices, socioeconomic conditions, and access to medical treatment. A state in eastern India, Jharkhand shows a remarkable frequency of both diabetes and dietary issues (11). Developing targeted treatment options calls for an understanding of how dietary patterns, the presence of pre-existing anaemia, bad behaviours, and other medical disorders common in this area interact with vitamin B12 deficiency and then influence haematological health in diabetes patients. There is limited specific data describing these intricate interactions inside the diabetes population of Jharkhand. The goal of this study is to unravel the interaction of diet, pre-existing anaemia, negative habits, and medical history with the degree of vitamin B12 deficit and their consequences for haematological health in a group of adult type 2 diabetes mellitus patients visiting a tertiary care centre in Jharkhand. Examining these links will help us to offer a more complete picture of the elements affecting B12 level and haematological characteristics in this particular and understudied group.

Methods

Study Design and Setting: This cross-sectional, observational study was conducted at the outpatient department of a tertiary care hospital in Jharkhand, India, over a period of Specify Study Duration, e.g., 09 months from January 2024 to September 2024. The hospital serves a diverse adult population with a significant burden of type 2 diabetes mellitus and varying nutritional profiles.

Study Population and Sampling; We enrolled a total of 420 adult patients (aged ≥ 18 years) diagnosed with type 2 diabetes mellitus (T2DM) based on the American Diabetes Association criteria (2023). Participants were consecutively recruited during their routine visits to the outpatient department.

Data Collection

After obtaining informed written consent, detailed information was collected from each participant using a structured questionnaire and review of medical records. This included:

- **Dietary Habits:** Categorized as vegetarian or non-vegetarian based on self-reported primary dietary pattern.
- **Presence of Hemoglobin Disorders (Anemia):** Documented history of anemia prior to the current presentation, as recorded in their medical records.
- **Adverse Habits:** Information on current smoking status (smoker/non-smoker) and alcohol consumption (consumer/non-consumer).
- **Medical History:** Presence of relevant comorbidities, including gastrointestinal disorders (e.g., atrophic gastritis, Crohn's disease, celiac disease) and autoimmune diseases (e.g., rheumatoid arthritis, lupus), as documented in their medical records.
- **Vitamin B12 Levels:** Fasting venous blood samples were collected, and serum vitamin B12 levels were measured using a commercially available chemiluminescence immunoassay (CLIA) kit. The severity of vitamin B12 deficiency was categorized based on established cut-off values . Mild (100-179 pg/mL) and Severe (<100 pg/mL)
- **Hematological Parameters:** Complete blood counts (CBC) were performed using an automated hematology analyzer, including hemoglobin (Hb), red blood cell (RBC) count, white blood cell (WBC) count, differential leukocyte count (DLC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and platelet count.

Statistical Analysis

Data were analyzed using JAMOVI v 2.6.26. Categorical variables were compared using chi-square tests to assess the association between diet, presence of anemia, adverse habits, medical history, and the

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severity of vitamin B12 deficiency. Continuous variables (hematological parameters) were compared across these categories using independent t-tests or ANOVA, as appropriate based on the number of groups and normality of data. Multivariate regression analyses (linear for continuous hematological outcomes, logistic for binary outcomes like presence of specific cytopenias) were performed to assess the independent effects of diet, pre-existing anemia, adverse habits, and medical history on vitamin B12 levels and hematological parameters, adjusting for potential confounders such as age, gender, duration of diabetes, and metformin use. A p-value of < 0.05 was considered statistically significant.

Results

Table 1 presents the baseline distribution of key characteristics within the study cohort of 420 participants. Regarding dietary habits, 166 (39.53%) participants reported following a vegetarian diet, while 254 (60.47%) were non-vegetarians. A significant proportion of the cohort (53.81%) had a pre-existing history of anemia. In terms of adverse habits, the majority were non-smokers (88.10%) and non-alcohol consumers (95.00%). Concerning relevant medical history, 44 (10.50%) participants had a documented presence of a gastrointestinal disorder consistent with malabsorption syndrome, while 16 (3.80%) had a presence of other associated nutritional deficiencies.

Table 1: Distribution of Study Participants by Diet, Pre-existing Anemia, Adverse Habits, and Medical History

Serial number	Characteristic	N (%)
1	**Dietary Habits**	
	Vegetarian	166, (39.53%)
	Non-Vegetarian	254 (60.47%)
2	**Pre-existing Anemia**	
	Yes	226, (53.81%)
	No	194,(46.22%)
3	**Adverse Habits**	
	Smoker	50(11.90%)
	Non-Smoker	370(88.10%)
	Alcohol Consumer	21(5.0%)
	Non-Alcohol Consumer	399(95.0%)
4	**Medical History (Any Relevant)**	
	Presence of GI Disorder (Malabsorptionsyndrome)	44(10.5%)
	Absence of GI Disorder (Malabsorptionsyndrome)	376(89.5%)
	Not Applicable	131(31.2%)
	Presence of Associated Deficinecy	16(3.8%)
	Absence of Associated Deficinecy	273(65.0%)

Table 2 examines the association between dietary habits, pre-existing anemia, adverse habits, relevant medical history, and the severity of vitamin B12 deficiency, categorized as Mild (100-179 pg/mL) and Severe (<100 pg/mL).

No statistically significant association was observed between dietary habits (vegetarian vs. non-vegetarian) and the severity of vitamin B12 deficiency (p > 0.05).

A statistically significant association was found between pre-existing anemia and the severity of vitamin B12 deficiency (p < 0.05). A higher proportion of participants with pre-existing anemia (73.4%) fell into the mild deficiency category compared to those without pre-existing anemia (26.6%). Conversely, a smaller proportion of participants with pre-existing anemia (70.9%) were in the severe deficiency category compared to those without (29.1%).

Smoking habits also showed a statistically significant association with vitamin B12 deficiency severity (p < 0.05). A higher proportion of smokers (14.2%) had mild deficiency compared to severe deficiency (7.9%), while a higher proportion of non-smokers (92.1%) were in the severe deficiency group compared to smokers (85.8%). Alcohol consumption did not show a significant association with B12 deficiency severity (p > 0.05).

Regarding medical history of malabsorption syndrome, no statistically significant association was found with the severity of vitamin B12 deficiency (p > 0.05). However, a statistically significant association was observed for the presence of other associated deficiencies (p < 0.05). A higher proportion of participants with associated deficiencies (5.9%) fell into the severe deficiency category compared to those without (2.6%). Conversely, a smaller proportion of participants with associated deficiencies (2.6%) were in

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the mild deficiency category compared to those without (63.1%).

These findings suggest potential interactions between pre-existing anemia, smoking habits, presence of other associated deficiencies, and the severity of vitamin B12 deficiency in this cohort.

Table 2: Association of Diet, Pre-existing Anemia, Adverse Habits, and Medical History with Severity of Vitamin B12 Deficiency

Characteristic	Mild Deficiency (100-179 pg/mL) N (%)	Severe Deficiency (<100 pg/mL) N (%)	p-value (Chi-Square)
1. Dietary Habits			
Vegetarian	106 (39.6%)	55 (36.2%)	>0.05
Non-Vegetarian	162 (60.4%)	97(63.8%)	
2. Pre-existing Anemia			
Yes	193(73.4%)	107 (70.9%)	<0.05
No	70(26.6%)	44 (29.1%)	
3. Habits			
Smoker	38 (14.2%)	12 (7.9%)	<0.05
Non-Smoker	230 (85.8%)	140 (92.1%)	
Alcohol Consumer	14(5.2%)	7(4.6%)	>0.05
Non-Alcohol Consumer	254 (94.8%)	145 (95.4%)	
4. Medical History (Any Relevant)			
Presence of GI Disorder (Malabsorption syndrome)	29 (10.8%)	15 (9.9%)	>0.05
Absence of GI Disorder (Malabsorption syndrome)	239 (89.2%)	137 (90.1%)	
Presence of Associated Deficiency	7 (2.6%)	9 (5.9%)	<0.05
Absence of Associated Deficiency	169 (63.1%)	104 (68.4%)	

Table 3: Comparison of Mean Hematological Parameters Across Categories of Severity of Vitamin B12, and Pre-existing Anemia.

Hematological Parameter	Deficient B12 (100-179 pg/mL) Mean ± SD	Severely Deficient B12 (<100 pg/mL) Mean ± SD	p-value (t-test)	Pre-existing Anemia (No) Mean ± SD	Pre-existing Anemia (Yes) Mean ± SD	p-value (t-test)
Hemoglobin (g/dL)	11.70±2.39	11.53 ± 2.38	>0.05	14.17 ± 0.95	10.57 ± 1.80	<.001
RBC Count (x10^6/µL)	4.19± 0.74	4.00 ± 0.89	>0.05	4.75 ± 0.65	3.86 ± 0.70	<.001
WBC Count (/µL)	8038 ± 3013	7830 ± 3036.66	>0.05	8144.74 ± 2316.13	7819.87 ± 3165.66	<.001
Neutrophils (%)	63.68 ± 12.10	65.28 ± 13.80	<0.05	59.60 ± 11.24	65.97 ± 12.78	<.05
Lymphocytes (%)	25.52 ± 10.36	24.25 ± 11.69	>0.05	28.88 ± 9.06	23.69 ± 11.11	<.001
Monocytes (%)	3.06 ± 3.04	3.11 ± 4.76	>0.05	3.78 ± 5.38	2.82 ± 2.90	<.05
Eosinophils (%)	7.03 ± 2.38	7.19 ± 5.87	>0.05	7.89 ± 6.64	6.77 ± 2.31	>0.05
Basophils (%)	0.52 ± 0.51	0.50 ± 0.50	>0.05	0.53 ± 0.50	0.51 ± 0.51	>0.05
MCV (fL)	88.74 ± 8.96	92.93 ± 14.42	<0.00	93.41 ± 8.34	88.92 ± 12.20	<.05
MCH (pg)	28.12 ± 3.91	30.95 ± 20.44	<.05	30.11 ± 3.20	28.73 ± 14.93	>0.05
MCHC (g/dL)	31.49± 1.35	31.45 ± 1.37	>0.05	32.19 ± 0.98	31.19 ± 1.37	<.001
Platelet Count (/µL)	178773.51± 72233.66	175652.63± 80619.17	>0.05	191938.60 ± 67882.03	172231.67 ± 77879.33	>0.05

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Table 3 compares mean hematological parameters by vitamin B12 deficiency severity and pre-existing anemia. Severely deficient participants exhibited significantly higher MCV and MCH, along with a higher neutrophil percentage, compared to the deficient group. Pre-existing anemia was associated with significantly lower hemoglobin, RBC count, lymphocyte count, monocyte count, and MCV, but higher neutrophil percentage and MCHC, compared to those without pre-existing anemia. No significant differences were observed for other hematological parameters between the B12 severity groups or the pre-existing anemia groups.

Discussion

This cross-sectional study investigated the interplay of diet, pre-existing anemia, adverse habits, and medical history with the severity of vitamin B12 deficiency and their implications for hematological health in 420 adult patients with type 2 diabetes mellitus in Jharkhand, India. We categorized participants based on dietary habits, pre-existing anemia, smoking and alcohol consumption, and presence of malabsorption syndrome or associated deficiencies. Our salient findings reveal that while dietary habits and malabsorption syndrome did not significantly correlate with B12 deficiency severity, pre-existing anemia and presence of other associated deficiencies were associated with a higher likelihood of severe B12 deficiency. Furthermore, severe B12 deficiency was linked to higher MCV and MCH. Pre-existing anemia was associated with a distinct hematological profile characterized by lower hemoglobin, RBC count, lymphocyte count, monocyte count, and MCV, but higher neutrophil percentage and MCHC. These findings highlight the complex interplay of nutritional status and comorbidities in shaping the hematological landscape of B12 deficiency in diabetic individuals in this region.

Comparing our findings with existing literature, the lack of a strong association between vegetarian diet and B12 deficiency severity in our diabetic cohort, despite the known link in general populations, might suggest that other factors, such as metformin use (12), or duration of diabetes (13), could be playing a more dominant role in this specific group. However, our overall prevalence of vegetarianism (39.53%) is substantial and likely contributes to the high prevalence of B12 deficiency observed consistent with findings in other Indian studies (6)

The significant association between pre-existing anemia and severe B12 deficiency suggests a potential synergistic effect where chronic anemia might exacerbate the depletion of B12 stores or vice versa (14). The distinct hematological profile observed in patients with pre-existing anemia, characterized by lower red cell indices alongside altered white cell differentials, underscores the complex interplay of underlying hematological disorders and B12 deficiency.

Our finding of a significant association between smoking and B12 deficiency severity, with a higher proportion of non-smokers in the severe deficiency group, is less commonly reported and warrants further investigation. Some studies suggest that smoking might affect nutrient absorption and metabolism (15), but the direction of the association with B12 levels is not always consistent.

The significant association between the presence of other associated deficiencies and severe B12 deficiency suggests that individuals with multiple nutritional deficits might be at a higher risk for more pronounced B12 depletion, potentially due to overall poor nutritional status or underlying malabsorption issues (16).Interestingly, a recent study by Mercantepe et. al . also explored the relationship between B12 levels and comorbidities in diabetes, although their specific focus on obesity adds another dimension to the interplay of metabolic and nutritional factors (10).

The link between severe B12 deficiency and higher MCV and MCH aligns with the classical understanding of B12 deficiency leading to macrocytic anemia (17). However, the absence of a significant difference in mean hemoglobin levels between the B12 severity groups suggests that macrocytosis might precede overt anemia in some cases.

Strengths: This study provides valuable, region-specific data on the interplay of diet and comorbidities with vitamin B12 deficiency in a diabetic cohort from an understudied region of India. The inclusion of various baseline characteristics and a detailed hematological profile offers a comprehensive assessment.

Limitations: The cross-sectional design limits our ability to establish temporal relationships and causality. Dietary intake was assessed by self-report, which may be subject to bias. We did not measure B12 metabolites like methylmalonic acid and homocysteine, which could provide a more sensitive assessment of B12 status.

Conclusion

In conclusion, our study in a diabetic cohort from Jharkhand reveals that pre-existing anemia and the presence of other associated deficiencies are linked to a higher likelihood of severe vitamin B12 deficiency. Severe B12 deficiency is associated with macrocytosis, while pre-existing anemia presents with a distinct hematological profile. These findings underscore the complex interplay between nutritional status, comorbidities, and B12 deficiency in diabetic individuals. A comprehensive assessment of dietary habits, pre-existing conditions, and lifestyle factors is crucial for identifying and managing vitamin B12

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deficiency in this vulnerable population to optimize hematological health.

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