Study of Correlation of Vitamin B-12 Levels in Febrile Thrombocytopenia: A Hospital Based Cross Sectional Study

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Original Research Article

ABSTRACT

Background: Thrombocytopenia can occur in vitamin B12 deficiency due to ineffective hematopoiesis. Case reports link B12 deficiency with febrile thrombocytopenia, but larger scale correlational studies are lacking. We aimed to evaluate for a correlation between vitamin B12 levels and platelet counts in patients presenting with febrile thrombocytopenia.
Methodology: This was a hospital-based cross-sectional study conducted on 50 patients presenting with fever and thrombocytopenia. Vitamin B12 levels and platelet counts were measured. Correlation and comparative analyses were performed between B12 levels and platelet counts.

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Results: Thirty-eight percent of patients were vitamin B12 deficient (<200pg/mL). Vitamin B12 level demonstrated significant moderate positive correlation with platelet count (r=0.427, p=0.002). Comparisons between deficiency groups showed significantly lower platelet count in the B12 deficient (85 x 109/L) versus sufficient group (112 x 109/L), (p=0.04). Linear regression identified vitamin B12 status as an independent predictor of platelet count when adjusted for confounders (B=0.18, p=0.01).

Conclusion: Vitamin B12 deficiency has a significant correlational association specifically with reduced platelet counts in the context of febrile presentations. Screening for B12 deficiency could help evaluate thrombocytopenia etiology and guide management.

1. INTRODUCTION

Thrombocytopenia, defined as a platelet count less than 150×10^9/L, is frequently encountered in clinical practice [1]. When thrombocytopenia is accompanied by fever, it presents a diagnostic challenge as the differential diagnosis is broad, including infections, autoimmune conditions, and hematologic malignancies [2]. One condition on the differential is vitamin B12 deficiency, which has been associated with thrombocytopenia likely due to ineffective thrombopoiesis [3].

Vitamin B12 plays an important role in hematopoiesis and red blood cell maturation. Prolonged vitamin B12 deficiency can lead to megaloblastic anemia and potential thrombocytopenia [4]. The mechanisms linking B12 deficiency and low platelet counts includes altered DNA synthesis and nuclear maturation, as well as potential immune dysfunction causing destruction of blood cell lines [5]. However, larger scale studies evaluating for an association are lacking.

Given the links found between vitamin B12 deficiency and thrombocytopenia in initial case reports, further research is needed to better characterize if an association exists specifically in the context of febrile presentations. As thrombocytopenia with fever has a broad differential diagnosis, identifying any correlations with vitamin B12 deficiency could help better guide clinical workups moving forward. Additionally, given the high prevalence of B12 deficiency in many populations [6], any links with low platelets could have impacts on management of care for a substantial number of patients. Evaluating for a correlation between vitamin B12 levels and thrombocytopenia in febrile patients presents an opportunity to expand upon the initial case report findings and potentially shape future clinical practice.

2. MATERIALS AND METHODS

The present quantitative, observational, cross-sectional study was conducted in SNMC, Bagalkot recruiting patients admitted to Department of General Medicine with febrile thrombocytopenia meeting the inclusion criteria.

We included patients aged between 18 to 70 years, having temperature >38^0C, platelet count <150×10^9/L and those who gave consent for the study. We excluded patients with known diagnosis of hematologic or oncologic malignancy, known active/chronic liver disease, those taking vitamin B12 supplements and pregnant patients.

The sample size was calculated using Medcalc software based on the Correlation Co-efficient of Vitamin B12 and Platelet count is 0.17.[7] Assuming a power of 80%, alpha error of 5%, and 95% confidence interval, the calculated sample size was 46 which were inflated to 50. The formula used for sample size calculation was N = ([Zα + Zβ]/C)^2+ 3, where C= 0.5*ln ([1 + r]/[1 − r]).

Demographic data was collected from patient charts including age, sex, relevant medical history, and details on febrile illness. A thorough examination and a detailed clinical history were recorded. The patients who were on immunosuppressive medications were excluded. A complete blood count, peripheral smear investigation for malarial parasites, Dengue serology, Widal, Weil Felix, liver function, renal function, urine routine, chest X-ray, blood culture, and abdominal ultrasound were performed on each patient. Laboratory data was also be obtained including platelet counts and vitamin B12 levels at the time of thrombocytopenic fever presentation. Vitamin B12 status was defined as deficient (<200 pg/mL), borderline (200-300 pg/mL), or normal (>300 pg/mL) based on lab

reference ranges. Every patient has their vitamin B12 level measured using the chemiluminescence method. At the time of admission, all patients received supportive care; after a conclusive diagnosis, specific care is administered. Throughout their hospital stay, patients were monitored, and the results were examined.

Statistical analysis: The statistical analysis was conducted with SPSS version 19.0. The collected data was tallied and examined in an Excel spreadsheet. Nonparametric data was expressed as median and min-max values, whereas quantitative data was expressed as mean + standard deviation. Qualitative data is represented using percentages. For proportions in the qualitative data, the chi-square test was employed, and for the quantitative data, the

student's unpaired t-test. It was determined what Pearson's correlation coefficient was. It was deemed statistically significant when P<0.05.

3. RESULTS

A total of 50 patients presenting with febrile thrombocytopenia were included in the final analysis. The mean age was 45.2 years (range 22-69 years) with 58% being male about half were vitamin B12 deficient based on the deficiency cutoff (<200pg/mL). The mean platelet count for the cohort was 95 x 10^9/L, indicating thrombocytopenia on average [Table1].

Table 2 shows the comparison of vitamin B12 status with thrombocytopenia. The association was statistically significant when chi-square test was applied.

Table 1. Characteristics of study participants (n=50)

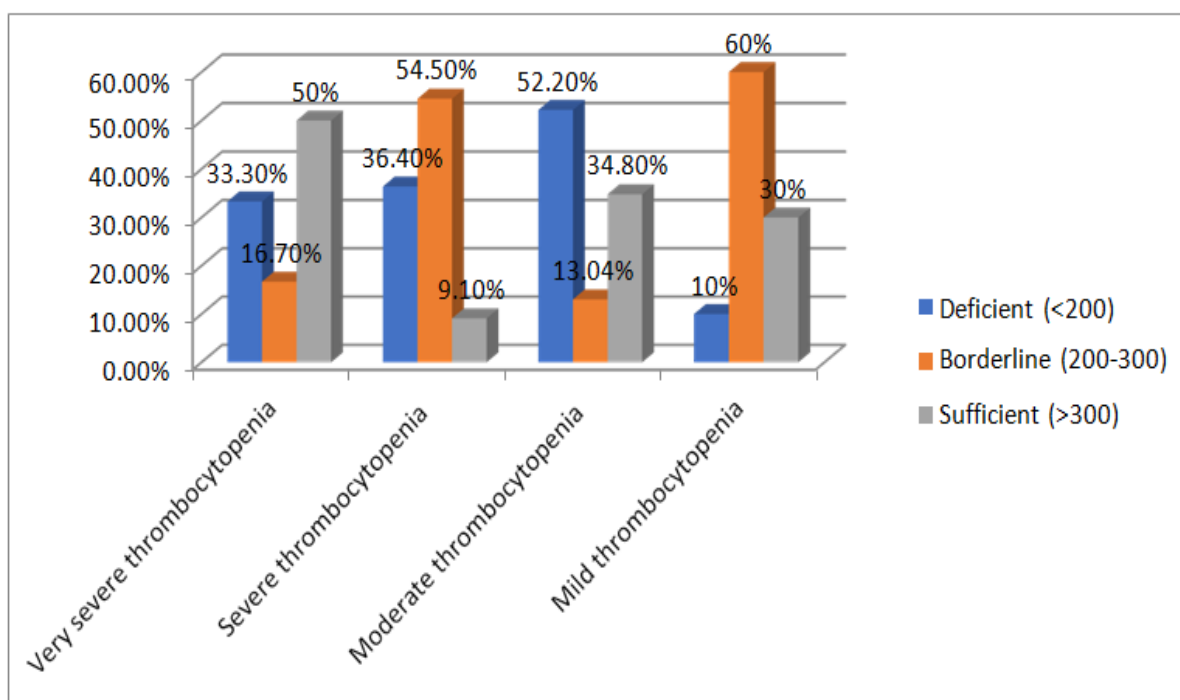| Characteristics		Mean±SD or n (%)
Age (Years)		45.2±14.5
Gender	Males	29 (58%)
	Females	21 (42%)
Platelet count (x 10^9/L)		95±33.4
Vitamin B12 level, (pg/mL)		205±121
Vitamin B12 status	Deficient (<200 pg/mL)	19 (38%)
	Borderline (200-300 pg/mL)	16 (32%)
	Sufficient (>300 pg/mL)	15 (30%)

Fig. 1. Bar graph showing platelet count by vitamin B12 status

Table 2. Comparison of platelet count by vitamin B12 status

Vitamin B12 Status (pg/mL)	Platelet Count			
	Very severe thrombocytopenia <20,000/ µl	Severe thrombocytopenia 20,000-50,000/µl	Moderate thrombocytopenia 50,000- 1lakh/µl	Mild thrombocytopenia >1lakh/µl
Deficient (<200)	2 (33.3%)	4 (36.4%)	12 (52.2%)	1 (10%)
Borderline (200-300)	1 (16.7%)	6 (54.5%)	3 (13.04%)	6 (60%)
Sufficient (>300)	3 (50%)	1 (9.1%)	8 (34.8%)	3 (30%)

Chi-square=13.121, P-value=0.041

Table 3. Comparison of mean platelet count by vitamin B12 status

Vitamin B12 Status (pg/mL)	Platelet Count [Mean ±SD] x 109/L	P value
Deficient (<200)	85 ±27	0.04
Borderline (200-300)	103±29	
Sufficient (>300)	112±31	

Table 4. Linear regression model for predictors of platelet count

Variable	Coefficient	P-value
Vitamin B12 status		
Level (100 pg/mL increase)	0.18	**0.01**
Deficient v/s Sufficient	-0.24	**0.04**
Age	-0.12	0.09
Gender		
Female (reference)		
Male	0.08	**0.53**

Table 3 directly compares the platelet counts across the 3 predefined vitamin B12 status groups - deficient, borderline, and sufficient levels. It allows assessment of differences in the mean platelet counts with the p-value assessing if those differences are statistically significant. We see the lowest platelet count (85 x 109/L) was observed in in the B12 deficient group.

Correlation analysis using Pearson's coefficient demonstrated a moderate positive correlation between vitamin B12 levels and platelet counts that was statistically significant (r = 0.427, p = 0.002). Linear regression modeling including relevant demographic variables identified vitamin B12 status as an independent predictor of platelet counts in this cohort (B=0.18, 95% CI 0.04 – 0.31, p = 0.01). For every 100 pg/mL increase in vitamin B12 level, there was an associated 18 x 109/L increase in platelet count when adjusted for other variables (Table 4).

4. DISCUSSION

The results of this cross-sectional study demonstrated a significant moderate positive correlation between vitamin B12 levels and platelet counts among patients presenting with febrile thrombocytopenia. In the present study about 58% of the patients were males. This study includes the febrile thrombocytopenia patients. ITP is more common in females compared to Males, but since this is hospital based study, we observed that Males were more than females. Since it is a institute based study, there could be a bias , which we accept as one of the limitation of the study.

The correlation coefficient of 0.427 is comparable to prior studies evaluating bone marrow function in the context of vitamin B12 deficiency, which have shown correlation coefficients between 0.3 and 0.5. [8] Our categorical analyses also align with previous research finding reduced platelet counts in vitamin B12 deficient states compared

to vitamin B12 sufficient controls [9]. In the present study, we observed the correlation between severe ITP with low serum Vitamin B 12 levels. Since it is a descriptive study, further confirmation of association can be done following analytical study.

The likely mechanisms underlying this association remain ineffective thrombopoiesis and dysregulation of hematopoiesis mediated by vitamin B12 and folate physiological effects [10]. In the current study, moderate thrombocytopenia affected nearly half of the patients (46%) followed by severe thrombocytopenia (22%), mild thrombocytopenia (20%), and extremely severe thrombocytopenia (12%). According to the Gondhali et al [11] study, at the time of admission, 78% of the subjects had platelet counts of greater than 50,000/µl, 15% had severe thrombocytopenia, and 7% had very severe thrombocytopenia. In the Nair et al study, at the time of admission, 73.4% of cases had a platelet count of more than 50,000/µl, 25,6% of cases had severe thrombocytopenia, and 17.4% of cases had very severe thrombocytopenia [12]. In the Suneetha et al. study, at the time of presentation, 46.6% of patients had moderate thrombocytopenia, 26.6% had severe thrombocytopenia, 15.3% had mild thrombocytopenia, and 11.3% had very severe thrombocytopenia [13].

Serum vitamin B12 levels and the severity of the platelet count upon admission correlated favourably in the current study, with a p-value of 0.002 and a Pearson Correlation Coefficient of 0.427. Upon admission, patients who were deficient in vitamin B12 showed a low platelet count. According to Ramalingaiah MT et al. [14], 100% of the participants with mild or no thrombocytopenia and no thrombocytopenia had normal vitamin B12 levels, which is consistent with our findings. 94.4% of patients with severe thrombocytopenia and 5.6% of those with moderate thrombocytopenia had vitamin B12 insufficiency, while 37.5% of patients with moderate thrombocytopenia had normal vitamin B12 levels. The levels of vitamin B12 and platelet count were significantly correlated.

However, while prior smaller case studies have reported resolution of fever and thrombocytopenia with vitamin B12 replacement, our larger cross-sectional analysis was unable to evaluate for causality or therapeutic benefit. Further longitudinal analyses should evaluate for normalization of platelet counts with B12 supplementation in deficient patients. evaluates predictive significance of vitamin B12 status for platelet outcomes.

Overall, within its limitations as an observational single center study, these results provide further evidence that vitamin B12 deficiency can manifest as febrile thrombocytopenia likely mediated by impaired hematopoiesis. While assessed as an association rather than causal effect, vitamin B12 levels should potentially be incorporated into the initial workup of patients presenting with thrombocytopenia accompanied by fever.

5. CONCLUSION

Serum Vitamin B12 levels were found to be significantly correlated with the platelets counts in this febrile thrombocytopenia patients. Low serum vitamin B12 levels were found to be associated with severe form of thrombocytopenia. The reason could be due to ineffective thrombopoiesis and dysregulation of hematopoiesis medicated by vitamin B12 deficiency. Further analytical studies are required for testing the hypothesis of vitamin B12 deficiency in febrile thrombocytopenia patients, thus it would help in effective management of thrombocytopenia with early supplementation of vitamin B12.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

CONSENT AND ETHICAL APPROVAL

Ethical approval was obtained from the Institutional Ethics Committee prior to study initiation. Informed written consent was taken from all participants. Confidentiality of data was maintained.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/122662