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Assessment of Vitamin B12 Status and Associated Factors among Hemodialysis Patients in Western Libya: A Multicenter Study

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ABSTRACT

This study aimed to evaluate serum Vitamin B12 levels among hemodialysis patients in Western Libya, and to investigate the comprehensive association between these levels and various factors, including demographic data, clinical variables (such as dialysis duration and metformin use), dietary habits, and the prevalence of clinical symptoms like fatigue and mood changes. This cross-sectional study was conducted on 80 patients across three centers (Nalut, Zintan, and Tripoli) between June to December 2025. Serum Vitamin B12 was measured using Chemiluminescent Immunoassay (CLIA) on the Snibe Maglumi 800 analyzer. The results showed that 12.5% of patients had a deficiency, 51.2% had normal levels, and 36.3% showed elevated levels (>900 pg/mL). Dietary supplementation was the most significant factor linked to B12 levels (P = 0.000). Significant variations were found between the three centers (P = 0.020), with Kidney dialysis center at Nalut Central Hospital showing the lowest deficiency rates. Importantly, the results showed no statistically significant correlation between B12 levels and clinical symptoms or metformin use, suggesting that symptoms in these patients may stem from other uremic factors. In conclusion, although supplementation effectively reduces deficiency, a high percentage of patients exhibit laboratory hypervitaminosis. This elevation may suggest the presence of functional vitamin B12 deficiency, necessitating a more precise diagnostic approach.

Keywords: Renal Failure, Hemodialysis, Vitamin B12, Dietary Supplements, Functional Deficiency, Libya.

تقييم حالة فيتامين ب12 والعوامل المرتبطة به لدى مرضى غسيل الكلى في غرب ليبيا: دراسة متعددة المراكز

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ملخـــــص البحـــــث

هدفت هذه الدراسة إلى تقييم مستويات فيتامين ب12 في مصل الدم لدى مرضى غسيل الكلى في غرب ليبيا، ودراسة العلاقة الشاملة بين هذه المستويات وعوامل مختلفة، بما في ذلك البيانات الديموغرافية، والمتغيرات السريرية (مثل مدة غسيل الكلى واستخدام الميتفورمين)، والعادات الغذائية، وانتشار الأعراض السريرية كالتعب وتقلبات المزاج المنهجية. تم إجراء هذه الدراسة المقطعية على 80 مريضا في ثلاثة مراكز (نالوت، والزنتان، وطرابلس) خلال الفترة من يونيو إلى ديسمبر 2025. وقد تم قياس مستوى فيتامين ب12 في مصل الدم باستخدام المقايسة المناعية الكيميائية الضوئية

(CLIA) على جهاز Snibe Maglumi 800 . وأظهرت النتائج أن 12.5% من المرضى يعانون من نقص فيتامين ب12، و51.2% لديهم مستويات طبيعية، و36.3% لديهم مستويات مرتفعة (>900 بيكوغرام/مل). وكان تناول المكملات الغذائية العامل الأكثر أهمية المرتبط بمستويات فيتامين ب12 (= P0.000). لوحظت اختلافات كبيرة بين المراكز الثلاثة (= P0.020)، حيث أظهر مركز غسيل الكلى في مستشفى نالوت المركزي أدنى معدلات النقص. ومن المهم ذكره أن النتائج لم تظهر أي ارتباط ذي دلالة إحصائية بين مستويات فيتامين ب12 والأعراض السريرية أو استخدام الميتفورمين، مما يشير إلى أن الأعراض لدى هؤلاء المرضى قد تنجم عن عوامل أخرى مرتبطة باليوريا، الخلاصة: على الرغم من أن المكملات الغذائية تقلل النقص بفعالية، إلا أن نسبة كبيرة من المرضى لديهم فرط فيتامين ب 12 في التحاليل المخبرية. قد يشير ذلك إلى وجود نقص وظيفي في فيتامين ب12، مما يستدعي اتباع نهج تشخيصي أكثر دقة.

الكلمات المفتاحية: الفشل الكلوي، غسيل الكلى، فيتامين ب 12، المكملات الغذائية، النقص الوظيفي، ليبيا.

1. Introduction

Vitamin B12 (cobalamin) is an essential nutrient vital for basic body functions, particularly the production of red blood cells, DNA synthesis, and keeping the nervous system healthy. Since the body cannot make this vitamin and must get it from food through a complex process, any problem in this process leads to a deficiency. This is very common in people with long-term illnesses, especially chronic kidney disease (CKD) [1]. The main issue is that a lack of Vitamin B12 in kidney patients does not only cause blood problems like anemia but also leads to nerve damage and memory issues. It is also linked to high levels of "homocysteine" in the blood, which increases the risk of heart disease, the top cause of death among kidney patients [2,3].

Research shows that patients on dialysis face extra risks; the dialysis process itself can wash the vitamin out of the blood, and the strict diets these patients follow make it harder to get enough B12 [4]. While only 1.5% to 15% of the general public suffers from this deficiency, it can reach up to 59% in kidney patients, especially those who have been on dialysis for a long time [5,6]. Other factors, like using the drug "metformin" or being elderly, also play a role [7]. On the other hand, some patients might have excessively high levels of B12 because of taking too many supplements or because their kidneys cannot clear certain proteins. These high levels can sometimes be misleading and may lead to serious health complications [8].

This study aims to fill the gap in information regarding Vitamin B12 levels among dialysis patients in Libya, where local data is limited. The goal is to measure how common B12 deficiency (or excess) is and to see how it relates to factors like age, gender, the length of time on dialysis, and the use of supplements or metformin. We hope these results will help doctors create better treatment plans to balance vitamin levels, improve patients' quality of life, and reduce complications.

2. Materials and Methods

2.1 Study Design

A cross-sectional, descriptive-analytical study was conducted between June and December 2025. The study targeted hemodialysis patients in three centers across Western Libya: Nalut Central Hospital, Zintan Dialysis Center, and Tripoli Kidney Services Center.

2.2 Participants and Sampling

A consecutive sampling method was used to recruit 80 adult patients (≥18 years). Participants were distributed as follows: Nalut (n=32), Zintan (n=31), and Tripoli (n=17). Patients who have been on dialysis for less than a year, or those who declined to participate were excluded.

2.3 Data Collection and Study Tools

Data were collected using a standardized questionnaire administered through interviews. The survey captured demographic characteristics (age and gender) and clinical factors, including the duration of hemodialysis, metformin use, dietary supplements, and nutritional habits, along with clinical symptoms.

2.4 Laboratory Analysis

Blood samples were collected pre-dialysis and prior to heparin administration to ensure the accuracy of serum Vitamin B12 measurements. Blood samples were collected into anticoagulant-free tubes and allowed to clot for 30 minutes at room temperature. The serum was then separated via centrifugation at 3000 rpm for 10 minutes. Serum Vitamin B12 levels were measured using a Chemiluminescent Immunoassay (CLIA) on the Snibe Maglumi 800 automated analyzer. Vitamin B12 status was categorized into three groups based on clinical standards: deficiency (<200 pg/mL), normal (200–900 pg/mL), and excess (>900 pg/mL).

2.5 Ethical Considerations

The study followed ethical research standards. Administrative approvals were obtained from the participating centers, and all patients gave their verbal consent. Participant privacy was protected using a serial numbering system, and the study involved no financial costs or health risks to the patients.

2.6 Statistical Analysis

Data were analyzed using SPSS (version 25). Descriptive statistics, including frequencies and percentages, were used to summarize baseline characteristics and Vitamin B12 status. The Chi-square test examined associations between Vitamin B12 levels and gender, supplement intake, metformin use, and clinical symptoms. Fisher's Exact Test was applied for age, duration of hemodialysis, diet, and comparisons between the three centers. Statistical significance was set at ($P < 0.05$).

3. Results

Table 1: Characteristics of the Study Population (n=80)

Variable	Category	Frequency (n)	Percentage (%)
Gender	Male	46	57.5%
	Female	34	42.5%
Age (Years)	< 40	22	27.5%
	40 – 60	46	57.5%
	> 60	12	15.0%
Dialysis Duration	< 3 Years	32	40.0%
	3 - 5 Years	18	22.5%
	> 5 Years	30	37.5%

The study included 80 patients. The majority were male (57.5%) compared to females (42.5%). Most participants (57.5%) belonged to the middle age group (40–60 years). In terms of dialysis

duration, 40% of patients had been on dialysis for less than 3 years, while 37.5% had been on treatment for more than 5 years.

Table 2: Vitamin B12 Status and Associated Factors (n=80)

Variable	Total (n)	Normal %	Deficient %	High %	X^2	df	P-value
Overall Prevalence	80	51.2	12.5	36.3	-	-	--
Gender (Male/Female)	46 / 34	54 / 47	11 / 15	35 / 38	0.497	2	0.780
Age (<40 / 40-60 / >60)	22/46/12	55/46/67	18/11/8	27/44/25	3.150	-	0.533
B12 Supplements - Yes / No	55 / 25	37 / 60	4 / 32	49 / 8	19.611	2	0.000*
Center Location - Nalut / Tripoli / Zintan	32/17/31	59.4 / 70.6 / 32.3	0 / 5.9 / 29	40.6/23.5 /38.7	15.618	-	0.002*
Metformin Use (Yes/No)	22 / 58	41 / 55	5 / 16	55 / 29	4.971	2	0.083
Dialysis Duration (1-3 / 3-5 / >5) Years	32/18/30	50/44/57	22/11/3	28/44/40	5.542	-	0.227
Dietary Adherence - Follows / No / Not Req.	44/32/4	55/41/100	11/16/0	34/44/0	4.475	-	0.318

Table 2 shows how Vitamin B12 levels are distributed among patients based on various factors. Statistical significance ($P < 0.05$) was found for two main factors: B12 supplement intake (P=0.000) and medical center location (P=0.020).

- B12 Supplements: Patients who did not take supplements had a much higher rate of deficiency (32%) compared to those who took them (4%). On the other hand, nearly half of the supplement users (49%) showed high Vitamin B12 levels. Statistical analysis confirmed a highly significant association for this factor ($X^2 = 19.611$, df = 2, P< 0.000).
- Center Location: Zintan General Hospital recorded the highest deficiency rate at 29%, while Nalut Central Hospital had no cases of deficiency (0%). Fisher's Exact Test revealed a highly significant clinical variation across the locations (Fisher's Exact Value = 15.618, P = 0.002).Other Factors: No significant statistical association was found between Vitamin B12 status and other variables, including gender $X^2 = 0.497$, df = 2, $P = 0.780$), age (Fisher's Exact Value = 3.150, P = 0.533), metformin use ($X^2 = 4.971$, df = 2, P = 0.083), duration of hemodialysis (Fisher's Exact Value = 5.542, P = 0.227), or dietary adherence (Fisher's Exact Value = 4.475, P = 0.318).

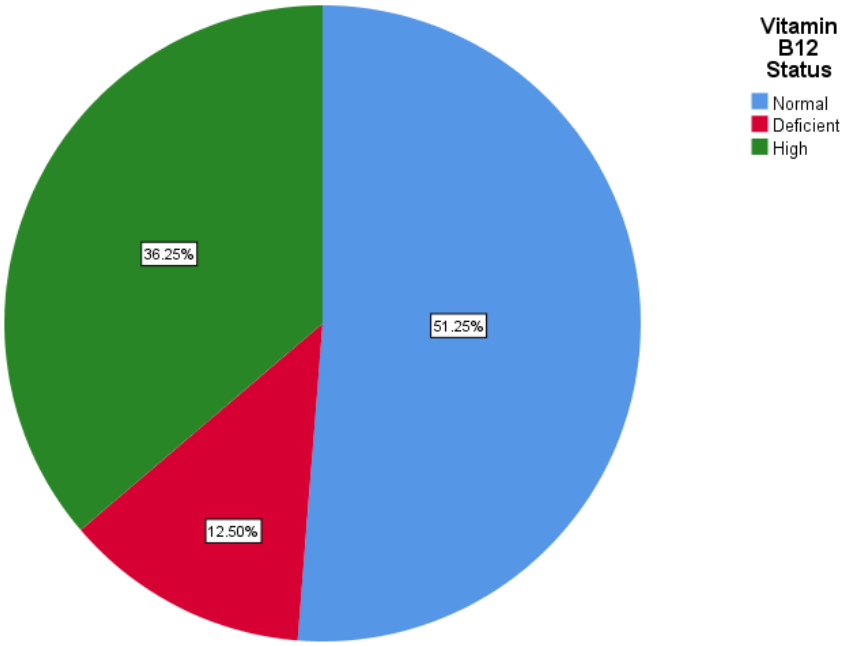

Figure 1: Distribution of Vitamin B12 levels within the study population

Figure 1 illustrates the overall prevalence of Vitamin B12 status among the study population (n=80). The results indicate that 51.25% of patients maintained normal B12 levels. Conversely, 12.50% were found to be deficient, while a significant portion (36.25%) exhibited high levels of the vitamin.

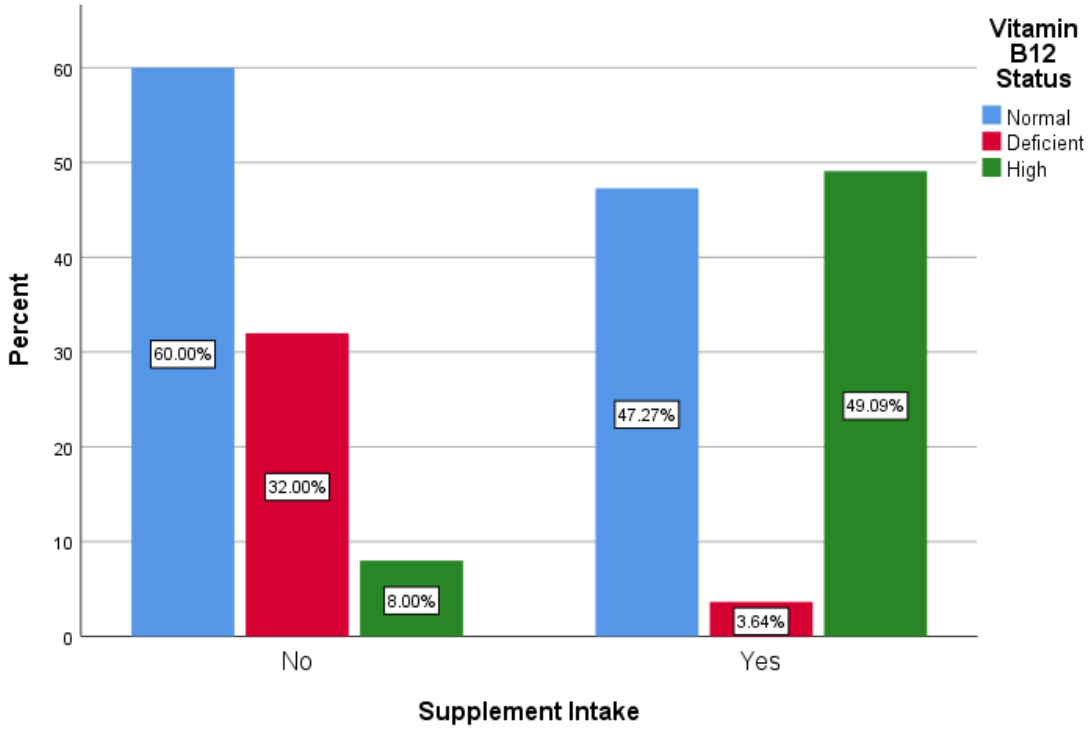

Figure 2: Distribution of Vitamin B12 levels according to supplement intake

As shown in Figure 2, the distribution of Vitamin B12 levels varies according to supplement intake. Statistical significance was highly evident (P=0.000), with a 32% deficiency rate in the non-supplement group compared to only 4% in the supplement group.

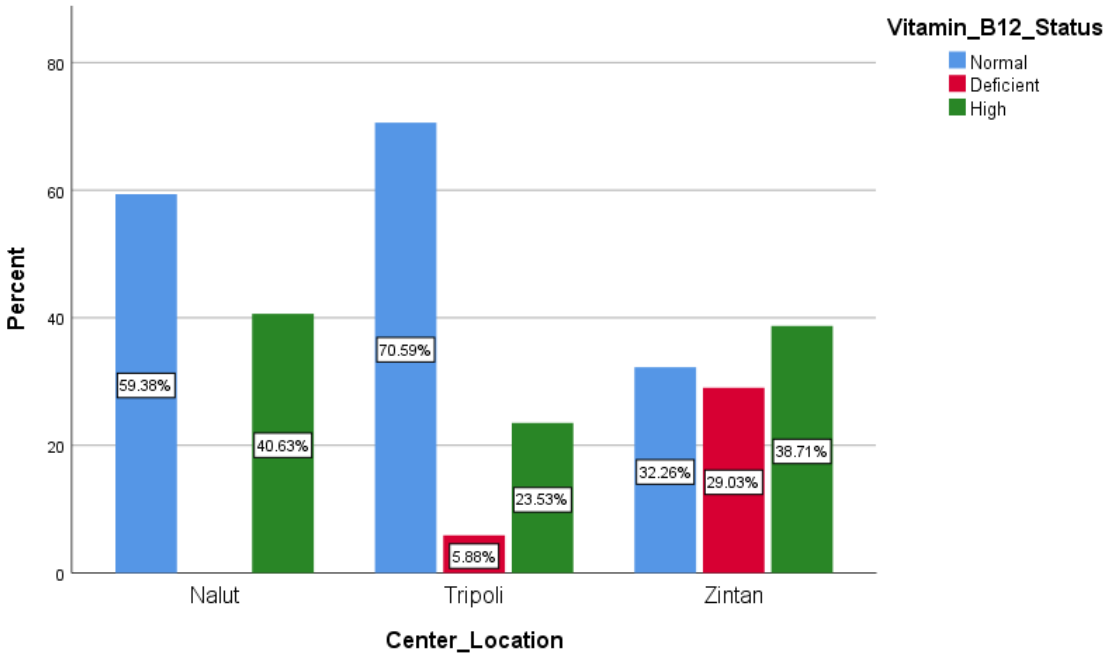

Figure 3: the variation in Vitamin B12 status across the three medical centers

Figure 3 illustrates the variation in Vitamin B12 status across the three medical centers. The results show statistically significant differences (P=0.002), with Zintan Center recording the highest deficiency rate at 29%, while Tripoli reported a low rate of 6%, and no deficiency cases were observed in Nalut (0%).

Table 3: Clinical Symptoms

Clinical Symptom	Prevalence n (%)	X^2	df	P-value (with B12 levels)
Fatigue	42 (52.5%)	1.089	2	0.580
Mood Swings	43 (53.8%)	1.846	2	0.397
Lack of Balance	30 (37.5%)	0.283	2	0.868
Limb Numbness	28 (35.0%)	3.042	2	0.218
Foot Swelling	22 (27.5%)	2.255	2	0.324
Memory Impairment	21 (26.3%)	0.173	2	0.917

The most frequent clinical symptoms reported by patients were mood swings (53.8%) and fatigue (52.5%). Other symptoms included lack of balance (37.5%) and limb numbness (35.0%). Despite their prevalence, statistical analysis showed that none of these symptoms were significantly associated with serum Vitamin B12 levels (P > 0.05), Specifically, no statistically significant association was observed for:

- Fatigue: (X^2 = 1.089, df = 2, P = 0.580)
- Mood Swings: (X^2 = 1.846, df = 2, P = 0.397)
- Lack of Balance: (X^2 = 0.283, df = 2, P = 0.868)
- Limb Numbness: (X^2 = 3.042, df = 2, P = 0.218)
- Foot Swelling: (X^2 = 2.255, df = 2, P = 0.324)
- Memory Impairment: (X^2 = 0.173, df = 2, P = 0.917)

These findings suggest that these clinical symptoms in hemodialysis patients may be attributed to other secondary clinical complications (such as uremic toxins, anemia, or electrolyte imbalances) rather than being solely driven by Vitamin B12 status.

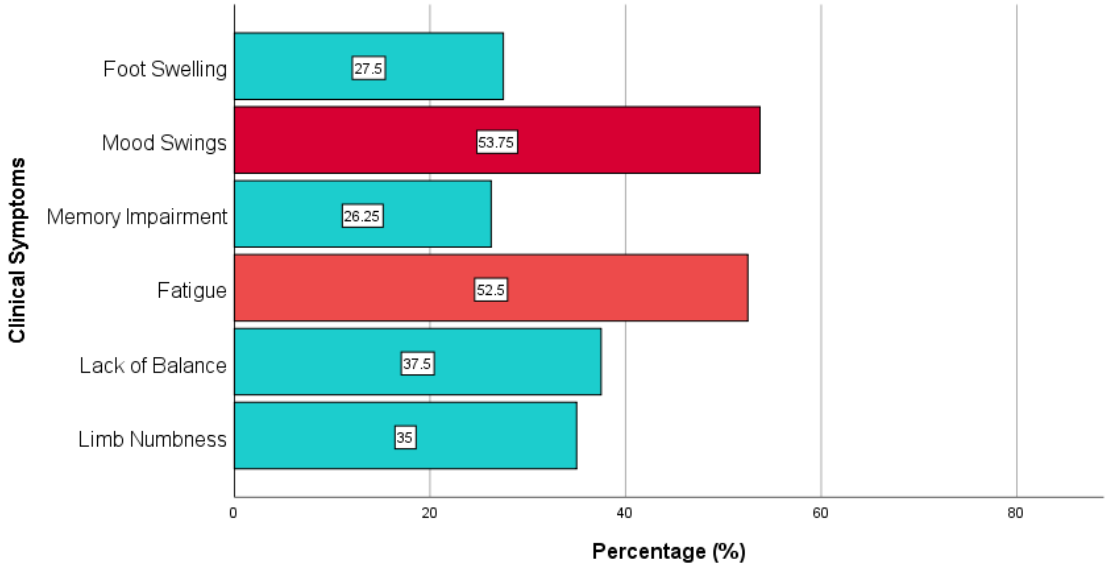

Figure 4: Prevalence of clinical symptoms among the study population

The chart visually represents the clinical symptom prevalence among the study group (n=80), clearly highlighting the high frequency of mood swings (53.8%) and fatigue (52.5%).

4. Discussion

4.1 Overall Prevalence of Vitamin B12 Status

The study found that the prevalence of Vitamin B12 deficiency was 12.5%. This is significantly lower than regional findings, such as the 73% reported in Iraq [9] and 58.8% in India [1]. This low deficiency rate reflects the effectiveness of B12 supplementation programs in the studied centers.

On the other hand, the study recorded a high prevalence of hypervitaminosis B12 (36.3%). This is likely an indicator of functional B12 deficiency in hemodialysis patients. This phenomenon occurs because most of the vitamin binds to Haptocorrin, a carrier protein that lacks cellular receptors in most tissues, making the vitamin biologically unavailable [10]. In healthy individuals, the liver clears this complex or excretes it via bile. However, in renal failure, impaired kidney function and saturated hepatic pathways lead to the accumulation of Haptocorrin and vitamin analogues. This raises serum levels in laboratory tests without reflecting actual cellular adequacy [11].

To distinguish between true and false elevations, it is recommended to use functional markers like Methylmalonic Acid (MMA) and Homocysteine. Elevated levels of these markers, despite high serum B12, confirm functional deficiency due to poor cellular uptake or interference from uremic toxins [12].

4.2 Demographic and Geographical Factors

Results showed no statistical significance between B12 levels and gender (P=0.780) or age (P=0.533). This suggests that the risk of B12 imbalance in Libyan hemodialysis patients is a

general risk regardless of age or sex, aligning with previous research [7], which stated that renal status itself is the primary determinant of vitamin levels. However, geographical location emerged as a significant factor (P=0.002), with notable variations between centers. Zintan General Hospital recorded the highest deficiency rate (29%), while Nalut Central Hospital showed no cases of deficiency (0%). This discrepancy suggests that local factors such as variations in clinical management protocols, differences in dietary habits across regions, or the availability of nutritional support may play a role.

4.3 Clinical Factors

Regarding dialysis duration, no significant correlation was found (P=0.227). This contradicts some studies suggesting that longer dialysis history increases vitamin depletion [13]. This discrepancy indicates that consistent supplementation in our sample successfully broke the link between disease duration and deficiency. As for Metformin, although it showed a slight influence (P=0.083), it did not reach statistical significance. This suggests that its effect might be more evident in larger samples or with higher doses, as noted in other regional research [14].

4.4 Dietary Factors

The results confirmed that supplementation is the most influential factor (P=0.000). The deficiency rate dropped from 32% in those not taking supplements to only 4% in those who were. This confirms that supplements are the primary line of defense. Conversely, dietary adherence did not show a significant impact (P=0.318), reinforcing the hypothesis that diet alone is insufficient to correct deficiency in these patients without medical intervention [4].

4.5 Clinical Symptoms

Despite the high prevalence of mood swings (53.8%) and fatigue (52.5%), no statistical correlation was found with B12 levels ($P > 0.05$). This suggests that such symptoms in hemodialysis patients are non-specific and may result from uremic toxins, anemia, or mineral imbalances rather than B12 deficiency alone. This differs from non-renal patients, where deficiency is more directly linked to neurological symptoms [15].

5. Conclusions

In conclusion, this study demonstrates that while Vitamin B12 deficiency is relatively well managed in the surveyed Libyan dialysis centers due to supplementation, there is a significant phenomenon of elevated B12 levels (36.3%). This elevation is often misleading as it may not reflect actual cellular availability due to the accumulation of inactive haptocorrin in renal failure. Therefore, this study strongly recommends that clinical assessment should not rely solely on total serum B12. Instead, it is crucial to incorporate functional biomarkers, specifically Homocysteine and Methylmalonic Acid (MMA), to accurately diagnose functional deficiency and optimize supplementation protocols for hemodialysis patients.

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Study Limitations

Small Sample Size: The study included only 80 patients, which may not represent all hemodialysis patients in Libya.

Study Design: Being a cross-sectional study, it cannot track changes in B12 levels over time or prove direct causes.

Lack of Functional Tests: The absence of MMA and Homocysteine measurements makes it difficult to confirm if high B12 levels mean the body is actually using it.

Self-reported Symptoms: Depending on patients own descriptions of their symptoms may lead to inaccurate or subjective results.

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