ARAŞTIRMA MAKALESİ / *RESEARCH ARTICLE*

Vitamin D, Vitamin B12 and Ferritin Levels in Children Presenting with Malnutrition Complaints

Malnütrisyon Şikâyetiyle Başvuran Çocuklarda D Vitamini, Vitamin B12 ve Ferritin Düzeyleri

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ABSTRACT

Aim: *In this study, it is aimed to examine the levels of [25(OH)D], vitamin B12 and ferritin, which we think are associated with the disease, in children who apply with the complaint of protein and energy deficiency.*

Material and Method: *The [25(OH)D] level of 1377 people who applied to the hospital, the vitamin B12 level of 1366 people and the ferritin level of 1384 people who applied to the hospital were included in the study. Hospital references for children were used for vitamin D [25(OH)D], vitamin B12 and Ferritin levels.*

Results: *When [25(OH)D] levels were examined in the study; Approximately 27% of the children had vitamin D deficiency [25(OH)D] <20 ng/mL, 7.19% had excessive [25(OH)D] deficiency and 6.68% had a high level of vitamin D deficiency. When vitamin B12 levels were examined, Vitamin B12 levels were low in 5.2% of the children (B12<191 ng/L) and high in 14.35%. Considering the ferritin levels, ferritin levels were low in 49.35% of children (ferritin level <30 µg/L).*

Conclusion: *It was observed that B12, [25(OH)D] and ferritin levels of patients with PEM could vary according to geographical regions. For this reason, we think it is necessary to focus on the age range of 6–24 months, where the incidence of micronutrient deficiencies and infectious diseases is the highest.*

Key words: *PEM; child; vitamin D; vitamin B12; ferritin*

ÖZET

Amaç: *Bu çalışmada protein ve enerji yetersizliği şikâyetiyle başvuran çocuklarda hastalıkla ilişkili olduğunu düşündüğümüz [25(OH)D], vitamin B12 ve ferritin düzeylerinin incelemesi amaçlanmaktadır.*

Materyal ve Metot: *Hastaneye başvuran 1377 kişinin [25(OH)D] düzeyi, 1366 kişinin vitamin B12 düzeyi ve hastaneye başvuran 1384 kişinin ferritin düzeyi çalışmaya dâhil edildi. D vitamini [25(OH)D], vitamin B12 ve Ferritin düzeyleri için hastanenin çocuklar için belirlediği referanslar kullanıldı.*

Bulgular: *Çalışmada [25(OH)D] düzeyleri incelendiğinde; Çocukların yaklaşık %27'sinde D vitamini eksikliği [25(OH)D] <20 ng/mL, %7,19'unda aşırı [25(OH)D] eksikliği ve %6,68'inde yüksek düzeyde D vitamini eksikliği saptanmıştır. B12 vitamin düzeyleri incelendiğinde; B12 vitamini düzeyi çocukların %5,2'sinde düşük (B12<191 ng/L), %14,35'inde yüksekti. Ferritin seviyelerine bakıldığında; Çocukların %49,35'inde ferritin seviyeleri düşüktü (ferritin seviyesi <30 µg/L).*

Sonuç: *Protein ve enerji yetersizliği (PEM) hastalarının B12, [25(OH)D] ve ferritin düzeylerinin coğrafi bölgelere göre değişebildiği görüldü. Bu nedenle mikrobesin eksikliklerinin ve bulaşıcı hastalıkların görülme sıklığının en yüksek olduğu 6–24 ay yaş aralığına odaklanılması gerektiğini düşünüyoruz.*

Anahtar kelimeler: *PEM; çocuk; D vitamini; B12 vitamini; ferritin*

Introduction

It is defined as protein-energy malnutrition (PEM) in the clinical-pathological picture that occurs in case of insufficient intake of one or more nutrients in a way that disrupts the body balance[1]. Childhood malnutrition accounts for approximately half (45%) of all deaths among children under the five worldwide[2]. Growth in children; It is an increase in body volume and mass with an increase in cell number and size. On the other hand, the development expresses bodily maturation with the change of cell, tissue and structure content[3]. The malnutrition rate is still high in developing countries[4]. Malnutrition affects one in nine people worldwide, and this rate seems to increase even more in low-income countries, especially among children younger than five years old. Malnutrition in early childhood can have harmful neurodevelopmental effects, with significant

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increases in lifelong cognitive, neurological, and mental health problems, and its consequences may spread to future generations[5]. 25-Hydroxy Vitamin D [25(OH) D] is a steroid vitamin that dissolves in adipose tissue. The most important effects are to keep calcium (Ca) and phosphor (P) levels in the normal physiological range along with parathyroid hormone (PTH) and thus to provide optimum bone mineralization[6].[25(OH)D] deficiency in children is one of the main causes of rickets. Rickets; Insufficient intake of [25(OH)D] occurs due to malabsorption, insufficient exposure to sunlight, and increased need during rapid growth, and the most common type is [25(OH)D] and/or caused by calcium deficiency. nutritional rickets[7]. Vitamin B12 deficiency in childhood is a rare disease with nonspecific symptoms. It was stated that many cases were exclusively breastfed and were children of mothers with low cobalamin levels. The development of the patients is normal in the early period. It is reported that symptoms and signs such as megaloblastic anemia, loss of acquired motor abilities, and growth and development retardation can be seen around 3–8 months[8]. Ferritin protein is found in body cells and especially in cells known as hepatocytes which make up approximately 75% of the liver, bone marrow and immune system cells. Ferritin, which is responsible for storing iron taken through food and releasing when necessary, is also defined as the body's iron store[9]. This study examines the levels of [25(OH)D], vitamin B12, and ferritin, which we think are associated with the disease, in children presenting with protein and energy deficiency complaints.

Materials and Methods

It was approved by a state University Ethics Committee (decision no: 2022/108, date: 21.09.2022). In the study, the results of vitamin B12, [25(OH)D] and ferritin levels requested from patients who applied to Health Sciences University Diyarbakir Gazi Yaşargil Training and Research Hospital between July 2020 and June 2022 were retrospectively analyzed. The reference range used by the hospital for children was taken as an example to diagnose vitamin B12, ferritin and vitamin D [25(OH) D] levels and to determine their normal ranges. Based on the reference ranges of 191–663 ng/L for Vitamin B12 and 30–400 μg/L for ferritin levels, results were grouped as low, normal and high levels. In addition, determining the degree of [25(OH)D] level <10 ng/mL severe deficiency, 10–19 ng/mL mild deficiency, 20–50 ng/mL normal, 51–80 ng An increased risk of hypercalciuria/ mL was assessed as >80 ng/mL of toxicity. The patients' data were obtained from electronic health records in the hospital database. Therefore, an informed consent form was not obtained from the patients. B12 levels of 1366 people, [25(OH)D] of 1377 people and ferritin levels of 1384 people who applied to the hospital were included in the study. B12, [25(OH)D] and ferritin levels were determined according to the age and gender of the patients. Patient names were kept confidential in the data analysis, and ethical rules were followed. The ages of children under the age of five included in the study were evaluated by dividing them into three groups: 6–24 months, 25–48 months and 49–60 months[10–11]. Serum B12, [25(OH)D], and ferritin levels were analyzed using the electrochemiluminescence method with a roche cobas device and an immunoassay system.

Statistical Analysis

Statistical Package for Social Sciences (SPSS) version 21.0 package program (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. The conformity of the data to the normal distribution was examined with the Kolmogorov-Smirnov test. Nonparametric tests were applied because they did not fit the normal distribution. The Mann-Whitney U test was used to determine whether there was a significant difference between children's Vitamin B12, [25(OH)D] and ferritin levels according to gender. Kruskal-Wallis test was used to determine whether the difference between the mean ages was significant. Frequency distributions, numbers, percentages, median, minimum and maximum values were given in descriptive statistics. A value of $P < 0.05$ was considered significant.

Results

The age and gender status of malnourished patients admitted to the hospital were divided into groups, and descriptive statistics, frequency distributions, and median, minimum and maximum values were given in the tables. Vitamin B12 and ferritin levels and [25(OH) D] vitamin levels are grouped according to the reference ranges used by the hospital for children, and the frequency distributions of low, normal and high level results by age and gender are given in Table 1. When [25(OH)D] levels were examined in this study, approximately 27% of the children (n=1377) had vitamin D deficiency (25(OH)D <20 ng/mL), of which 7.19% had excessive [25(OH)D] deficiency, and 6.68% had high levels of toxic [25(OH)D]. When children with [25(OH)D]deficiency are examined by gender; It has

Table 1. *Distribution of [25(OH)D], Vitamin B12 and Ferritin levels of children with malnutrition by age and gender*

	Age						Gender					
	6–24 month		25–48 month		49–60 month		Female		Male		Total	
	n	(%)*	n	(%)*	n	(%)*	n	(%)*	n	(%)*	n	(%)
[25(OH)D]												
<10 ng/mL	67	4.86 10	28	2.04 5.47	4	0.29 2.05	50	3.63 6.6	49	3.56 7.92	99	7.19
10–19 ng/mL	130	9.44 19.40	106	7.71 20.70	34	2.46 17.44	135	9.80 17.81	135	9.80 21.81	270	19.61
20–50 ng/mL	329	23.89 49.10	278	20.19 54.30	119	8.64 61.02	404	29.34 53.30	322	23.38 52.02	726	52.72
51–80 ng/mL	101	7.34 15.08	65	4.75 12.69	24	1.74 12.31	111	8.06 14.64	79	5.74 12.76	190	13.80
>80 ng/mL	43	3.13 6.42	35	2.51 6.84	14	1.01 7.18	58	4.21 7.65	34	2.47 5.49	92	6.68
Total	670	48.66 100	512	37.2 100	195	14.14 100	758	55.05 100	619	44.95 100	1377	100
Vitamin B12												
<191ng/L	52	3.81 7.76	11	0.80 2.18	8	0.59 4.19	33	2.42 4.37	38	2.78 6.23	71	5.20
191–663 ng/L	515	37.7 76.87	414	30.31 81.98	170	12.44 89.01	601	44 79.50	498	36.45 81.64	1099	80.45
>663 ng/L	103	7.54 15.37	80	5.86 15.84	13	0.95 6.80	122	8.93 16.13	74	5.42 12.13	196	14.35
Total	670	49.04 100	505	36.97 100	191	13.98 100	756	55.35 100	610	44.65 100	1366	100
Ferritin												
<30 µg/L	345	24.93 51.11	244	17.63 47.56	94	6.79 47.96	324	23.41 52.01	359	25.94 47.17	683	49.35
30–400 µg/L	330	23.84 48.89	269	19.44 52.44	102	7.37 52.04	299	21.60 47.99	402	29.05 52.83	701	50.65
>400 µg/L	0	0	0	0	0	0	0		0		0	0
Total	675	48.77 100	513	37.07 100	196	14.16 100	623	45.01 100	761	54.99 100	1384	100

* Values in the first row represent the percentage in the total, the values in the second row indicate the percentage in the group.

been determined that the rate of low levels in boys is higher than that of girls, and when looked at the age groups, it is seen at the highest rate of approximately 14% in 6–24-month-old children. There is a decrease in vitamin D[25(OH)D] deficiency rates as age progresses. When vitamin B12 levels were examined, they were found to be low (B12<191 ng/L) in 5.2% of children (1366), and high in 14.35%. When children with vitamin B12 deficiency are examined by gender, it is seen that the rates of low levels in girls are lower than in boys. When analyzed according to age groups, it is seen that the lowest rate is 3.81% in children aged 6–24 months. When the ferritin levels were examined, they were found to be low (ferritin level <30 µg/L) in 49.35% of children (n=1384). When the ferritin level was evaluated according to age, it was found that the lowest rate of 24.93% was seen in children aged 6–24 months, and there was a decrease in ferritin deficiency rates as age progressed. When the ferritin level is examined by gender, it has been determined that it is relatively lower in boys than in girls (male: 47.17%; Female 52.01%). The demographic information of the patients and the median, minimum and maximum levels of B12, [25(OH)D] and ferritin are given in Table 2. When the children under the age of five included in the study were examined according to their gender; For B12, the number of females was 756, the number of men was 610, for [25(OH)D] the number of women was 758, the number of men was 619, for ferritin the number of women was 623. The number of men was 761; the median B12 level was 384.5 ng/L in men, 409.5 ng/L in women. [25(OH)D] level was 31 ng/mL in females and 29 ng/mL in men, and ferritin level was 29 µg/L in women and 31 µg/L in men. The difference between the medians of B12 level, [25(OH)D] level and ferritin levels by gender was found to be statistically significant (p<0.05) (Table 2).

Table 2. *Evaluation of malnutrition patients by gender*

	Female			Male			p-value
	(n)	Median	Min-Max	(n)	Median	Min-Max	
B12(ng/L)	756	409.5	54–1759	610	384.5	78–1394	0.03
[25(OH)D] (ng/mL)	758	31	4–190	619	29	4–154	0.01
Ferritin (µg/L)	623	29	2–154	761	31	2–190	0.01

Table 3. *Evaluation of malnutrition patients by age*

	6–24 ay			25–48			49–60			P
	n	Median	Min-Max	n	Median	Min-Max	n	Median	Min-Max	
B12(ng/L)	670	384	54–1759	505	414	109–1527	191	382	88–1145	0.01
[25(OH)D] (ng/mL)	670	29	4–190	512	30.5	4–179	195	31	6–154	0.15
Ferritin (µg/L)	675	29	2–190	513	30	2–179	196	30.5	3–154	0.13

B12, [25(OH)D], and ferritin levels according to the age of the patients are given in Table 3. For B12, the number of patients aged 6–24 months was 670 median of 384 ng/L; the number of patients 25–48 months was 505 median of 414 ng/L; and the number of 49–60 patients was 191 median of 382 ng/L. The difference between the age groups of B12 levels according to the age of the children was found to be statistically significant (P <0.05). When paired comparisons were made, p=0.015 between 49–60 and 25–48, and p=0.004 between 6–24 and 25–48 were found to be significant. For [25(OH) D] vitamin, the number of patients 6–24 months old was 670 median of 29 ng/mL, the number of patients 25–48 months was 512 median 30.5 ng/mL, the number of 49–60 patients was 195 median 31 ng/mL. The difference between age groups was not statistically significant (p>0.05). For ferritin, the number of patients aged 6–24 months was 675 median of 29 µg/L; the number of patients 25–48 months old was 513 median of 30 µg/L; and the number of 49–60 patients was 196 median of 30.5 µg/L. The difference between age groups was not statistically significant (p>0.05) (Table 3).

Discussion

Protein-energy malnutrition (PEM) is one of the most common health problems in developing countries and mostly affects children between six months and five years[12]. Protein-energy malnutrition is seen at a rate of 11% to 69.5% in children aged 0–6 years in Türkiye, depending on different regions and settlement characteristics, and emerges as an important health problem[13,14]. Malnutrition kills more than 5 million children each year. In developing countries, 1–5% of children under five die from severe malnutrition[15]. Protein-energy malnutrition is common in developing countries and poor areas of developed countries. In this case, the most affected age group is infants and children under five[16,17]. Adequate nutrition in children; It is possible by taking and using the calories, protein, vitamins, minerals and trace elements necessary for maintaining life and adequate growth[18–19]. In a study by Nahide et al. with 134 patients, it was stated that 30.6% of children had [25(OH)D] deficiency[20]. Similarly, in a study conducted in Pakistan, it was stated that [25(OH)D] deficiency in children was 33.6%[21]. A study conducted in sub-Saharan Africa between 2012 and 2014 stated that the prevalence of [25(OH)D] in children is 28%[22]. In this study, the rate of [25(OH)D] deficiency in children was 27%. This rate was slightly below the literature rates. In a study conducted by Anver J et al. (2020) on 60 children with malnutrition, they stated that 18.3% of the children had low levels of vitamin B12 and these low rates were higher in boys[23]. Ng'eno et al. examined the vitamin B12 levels of 2166 children in 2017; 30.2% of the children reported low (vitamin B12 level <150 ng/L) vitamin B12 levels, which were lower in younger children and relatively higher in females than men, and stated that it was low at high rates[24]. In a study by Karagül and Yiğit (2022), in which they examined the vitamin B12 levels of 1874 children, they reported that vitamin B12 deficiency in children was 6.29%[25]. A study by Çolak et al. (2019) on 7310 children found the rate of children with vitamin B12 deficiency to be 16.9%[26]. In this study, it was found that vitamin B12 levels were low in 5.2% of the children, the miscarriage rates were higher in men than in women, and low rates were also higher in younger age groups. Vitamin B12 deficiency rates in the literature vary from country to country and even from region to region. It can be said that this is due to the nutritional characteristics of the studied populations and the different cut-off points of vitamin B12 levels in the laboratory.

Vitamin B12 deficiency rates in our study are similar to some studies in the literature but differ from other studies. These differences are due to the high cut-off point (B12<191 ng/L). According to age groups, it is similar to the literature, vitamin B12 deficiency in childhood is a rare disease with nonspecific symptoms. It has been reported that many cases are exclusively breastfed and children of mothers with low cobalamin levels[8]. Studies have been published reporting that malnutrition in the population may differ between genders. In a study by Şensoy et al., women's malnutrition rate was higher than that of men[27]. In our study, men's malnutrition rate in was higher than women's. This may be due to the difference in the study's number of patients and population. In a study on micronutrient deficiency among children in China, it was reported that there was no significant difference in B12 between genders[28]. A study conducted in India between 6–59 months of age reported that B12 deficiency is more pronounced in children under 24 months[29]. In our study, B12 was found to be 384 ng/L in children aged 6–24 months, 414 ng/L in children aged 25–48 months, and 382 ng/L in children aged 49–60 months. We think this is because children aged 6–24 months do not get enough B12 in their breast milk. In the 25–48 month old, it is seen that the child gets enough B12 from the foods he eats. However, low B12 again draws attention at 49–60 months of age. This draws attention to the fact that the child is at the age of 4–6 to start nursery or kindergarten, and does not get enough nutrition in this period. [25(OH)D] Vitamin deficiency is common in children and is accepted as a major health problem worldwide[30]. Walli et al.'s study on [25(OH)D] reported that 56.7% of malnourished people were men[20]. In our study, the level of [25(OH)D] vitamin in females was higher than in men and a statistically significant difference was found according to gender. Walli et al.'s study on [25(OH)D] reported that malnourishment was seen in children under two[20]. When the [25(OH)D] vitamin levels are evaluated according to age groups in our study, it is seen that it is 29 ng/mL in 6–24 months old, 30.5 ng/mL in 25–48 month old, 31 ng/mL in 49–60 month old. In our study, it is noteworthy that there is an increase in [25(OH)D] vitamin levels depending on age. It can be said that this increase is due to the increase in the duration of contact with the sun due to age progression. Serum ferritin level plays an important role in the diagnosis of iron deficiency. However, acute and chronic infections and liver disorders affect ferritin levels in PEM patients and act as acute phase reactants[31]. Saka et al. reported that ferritin levels were higher in female[32]. In our study, ferritin levels were found to be significantly lower in women compared to men. The reason for this difference in our study; It may have been caused by the population included in the study and the effect of geographical conditions. Wang et al.'s study of children under the age of three reported that the prevalence of anemic was higher in children under the age of two[33]. In our study, when the ferritin level was evaluated according to age, it was found to be 29 μg/L at 6–24 month old, 30 μg/L at 25–48 month old, 30.5 μg/L at 49–60 month old. In our study, it is noteworthy that there was an increase in the ferritin levels of the patients depending on age. It can be said that this is because adequate amounts of iron are not transferred with breast milk until babies are 24 months old, and it increases with additional food supplements at later ages.

Limitations of the Study

The main limitation of our study is that it was retrospective and the height and weight of the patients were not on the system. However, different results could have been obtained had the prospective study method been used.

Conclusion

In conclusion, B12, [25(OH)D], and ferritin levels of patients with PEM may vary according to geographical regions depending on regional dietary habits. Complementary nutrition interventions often focus on the 6–24 month age range, where growth stagnation, micronutrient deficiencies and the highest incidence of infectious diseases in developing countries, also need to consider gender situations. Therefore, effective interventions in reducing malnutrition should be a high priority. While the entire 6–60 month range is important, various interventions should be targeted in the first two years.

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Conflict of Interest

The authors declare they do not have any conflict of interest.

Author (s) Contribution

VT planned, conducted research work, and drafted the manuscript. ET guided the research.

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