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Vitamin D levels in patients with non-Hodgkin lymphoma/diffuse large B-cell lymphoma, chronic lymphocytic leukemia and multiple myeloma

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Abstract

Purpose: To investigate serum vitamin D levels in patients newly diagnosed with non-Hodgkin lymphoma/diffuse large B-cell lymphoma (NHL-DLBCL), multiple myeloma (MM) and chronic lymphocytic leukemia (CLL).

Patients and methods: We measured serum levels of vitamin D by ELISA in 103 patients prior to initiation of treatment, of whom 37 were diagnosed with MM, 32 with CLL and 34 with NHL-DLBCL.

Results: Suboptimal serum vitamin D levels (<30 ng/mL) were observed in all 103 patients. In 14 patients, serum vitamin D levels were between 20 and 30 ng/mL, while all other patients had vitamin D deficiency (<20 ng/mL). Severe vitamin D deficiency (<10 ng/mL) was observed in 32.3% of NHL-DLBCL patients, 28.1% of CLL patients and 81% of MM patients.

Conclusion: We observed low serum vitamin D levels in the majority of patients newly diagnosed with NHL-DLBCL, CLL and MM.

Keywords

Vitamin D, non-Hodgkin lymphoma, diffuse large B-cell lymphoma, multiple myeloma, chronic lymphocytic leukemia, serum biomarkers

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Introduction

Vitamin D began its evolutionary history more than a billion years ago as an inert molecule. Over time, it evolved into a

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hormone with multiple extracellular effects that regulate the expression of about 2000 genes.[1,2] 1,25-dihydroxyvitamin D or 1,25 (OH)2D exerts its biological activity by regulating gene transcription (genomic effects) and by activating signal transduction pathways near plasma membranes (non-genomic or rapid effects).[3] The mechanism of action involves binding to the vitamin D receptor, leading to activation or suppression of transcription.[4–6] Vitamin D deficiency is a global health problem. Globally, 1 in 7 people (14%) are thought to suffer from vitamin D insufficiency or deficiency.[2] In clinical practice, serum levels of 25(OH)D are used to evaluate vitamin D status in the body. Despite a lack of strong consensus on optimal levels of vitamin D, experts consider the lower limit of 30 ng/mL to be an adequate level (Table 1).[7] The suggested limit of 1,25(OH)2D for optimal skeletal health is a level that minimizes parathyroid hormone and maximizes calcium absorption.[8,9] Many studies have assessed potential links between vitamin D deficiency and the occurrence of neoplastic diseases as well as their outcomes. The purpose of the study was to investigate serum vitamin D levels in patients newly diagnosed with non-Hodgkin lymphoma/diffuse large B-cell lymphoma (NHL-DLBCL), multiple myeloma (MM), and chronic lymphocytic leukemia (CLL).

Table 1. Diagnostic levels of vitamin D.

1,25(OH)2D level (ng/mL)	1,25(OH)2D level (nmol/L)	Laboratory diagnosis
<10	<25	Severe deficiency
<20	<50	Deficiency
20–30	50–75	Insufficiency
30–100	75–250	Normal in sunny countries
>100	>250	Excess
>150	>325	Intoxication

Patients and methods

Patients

Vitamin D levels were assessed in patients with NHL-DLBCL, CLL and MM prior to treatment initiation. The patients were diagnosed at the Clinical Hematology Clinic of UMHAT St. George, Medical University of Plovdiv, from November 2014 to April 2016. The study was approved by the Scientific Ethics Committee of the Medical University of Plovdiv and was conducted in accordance with the principles laid out in the Helsinki declaration. All patients participating in the study signed an informed consent form. Patient data are presented in Table 2.

Statistical methods

Descriptive statistics and statistical inferences and conclusions were used to conduct the study. Data processing was carried out using IBM SPSS, version 25.0 (IBM Corp., Armonk, NY, USA) and MS Excel 2017 (Redmond, WA, USA). Statistical significance in testing the null hypothesis H_0 was assumed for values of $\alpha = 0.05$ (Z criterion = 1.96).

Measurement of vitamin D

Serum tests for vitamin D were performed at the first hospitalization of patients before starting treatment. Serum vitamin D levels were tested at the Central Clinical Laboratory at UMHAT St. George in Plovdiv, Bulgaria. An ELISA was used to quantify 1,25(OH)2D levels (Immundiagnostik, Bensheim, Germany; Catalog No. K2109).

Results

A total of 103 patients were enrolled in this study. Thirty-seven patients (19 women, 18 men) with a mean age of 68 years had a

Table 2. Patient characteristics.

Patients with MM (n = 32)		
Gender	Female	51.4%
	Male	48.6%
Mean age	68 years (38–86 years)	
Clinical stage (Durie & Salmon)	IA	10.8%
	IB	0%
	IIA	13.5%
	IIB	5.4%
	IIIA	18.9%
Clinical stage (ISS)	I	13.5%
	II	13.5%
	III	70.0%
Patients with NHL-DLBCL (n = 34)		
Gender	Female	47.1%
	Male	52.9%
Mean age	60.4 years (38–82 years)	
Clinical stage (Ann Arbor)	IA	11.8%
	IB	2.9%
	IIA	8.8%
	IIB	11.8%
	IIBs	2.9%
	IIIA	8.8%
	IIIB	23.5%
	IIIBs	2.9%
	IVB	14.7%
Extranodal involvement	Yes	44.1%
	No	55.9%
"B" symptoms	Yes	41.3%
	No	58.7%
Patients with CLL (n = 32)		
Gender	Female	34.4%
	Male	65.6%
Mean age	68 years (45–90 years)	
Clinical stage (Rai)	0	3.1%
	I	43.8%
	II	37.5%
	III	6.3%
	IV	9.4%
Clinical stage (Binet)	A	62.5%
	B	25.0%
	C	12.5%

diagnosis of MM according to the criteria of the International Myeloma Working Group. Thirty-two patients (11 women, 21 men) with a mean age of 68 years had a diagnosis of CLL according to the International Workshop on Chronic Lymphocytic Leukemia criteria. Thirty-four patients (16 women, 18 men) with a mean age of 60 years had a histologically confirmed diagnosis of NHL-DLBCL.

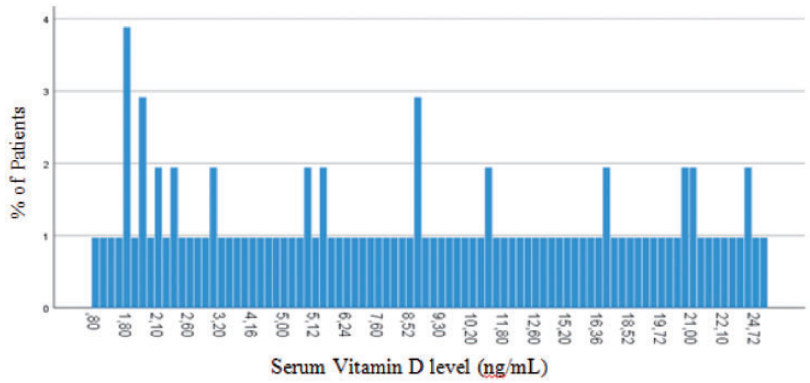

Figure 1. Level of vitamin D in all patients.

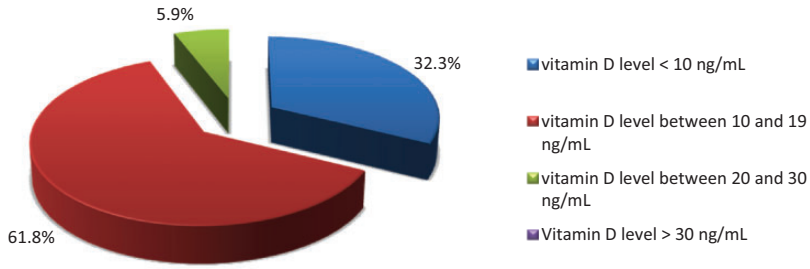

Figure 2. Distribution of patients with NHL-DLBCL according vitamin D level.

Serum levels of vitamin D below the optimum (<30 ng/mL) were observed in all 103 patients. The lowest measured level of vitamin D was 0.80 ng/mL and the highest measured level was 28.60 ng/mL. In 14.42% of patients, serum vitamin D levels were between 20 and 30 ng/mL, while all other patients had vitamin D deficiency (<20 ng/mL). These results are presented in Figure 1. Severe vitamin D deficiency (<10 ng/mL) was observed in 58.3% of patients and vitamin D levels between 10 and 20 ng/mL were observed in 27.28% of patients.

Vitamin D and NHL-DLBCL

In all 34 patients (17 men, 17 women) with newly diagnosed NHL-DLBCL, serum levels of vitamin D below the optimum (<30 ng/mL) were observed. Two patients had vitamin D insufficiency (serum levels between 20–30 ng/mL), while all other patients had vitamin D deficiency (serum levels <20 ng/mL). Severe vitamin D deficiency (<10 ng/mL) was observed in 61.8% of patients. These results are presented in Figure 2. No statistically significant differences were observed between serum vitamin D levels in men (9.3±5.7 mg/mL) and women (10.1±7.1 mg/mL). There was also no significant difference in vitamin D level associated with age. No statistically significant association was identified between vitamin D levels and Ann Arbor clinical stage. Patients with "B" symptoms had lower mean vitamin D levels. The Revised International Prognostic Index (R-IPI) and

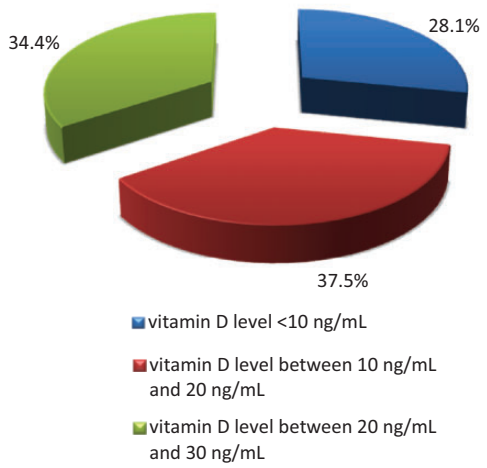

Figure 3. Distribution of patients with CLL according to vitamin D level.

National Comprehensive Cancer Network International Prognostic Index (NCCN IPI) were used to determine patient prognosis. A statistically significant association was identified between R-IPI and vitamin D levels ($p = 0.038$). Patients with lower levels of vitamin D had worse prognoses than patients with higher levels of vitamin D. No such correlation was identified between NCCN IPI and vitamin D levels. All patients were treated with rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP) or CHOP-like regimens as a first line therapy. Patients with the lowest levels of vitamin D had the worst responses to treatment and progressed most quickly.

Vitamin D and CLL

In all 32 (21 male, 11 female) patients with newly diagnosed CLL, suboptimal serum levels of vitamin D (<30 ng/mL) were observed. Eleven patients had vitamin D insufficiency (serum levels between 20–30 ng/mL), while the remaining 21 patients had vitamin D deficiency (levels <20 ng/mL). Severe vitamin D deficiency (<10 ng/mL) was observed in 28.1% of patients. These results are presented in Figure 3. No statistically significant difference was observed between serum vitamin D levels in men (15.4 ± 6.8 mg/mL) and women (13.6 ± 9.3 mg/mL). There was also no significant difference in vitamin D level associated with age. A statistically significant association was identified between vitamin D level and clinical stage according to the Rai and Binet systems. Patients with higher levels of vitamin D had lower clinical stages and less often required chemotherapy. Patients with higher leukocyte counts were found to have lower levels of vitamin D ($p = 0.017$). A similar relationship was found observed serum vitamin D levels and serum lactate dehydrogenase (LDH) levels. Patients with higher levels of vitamin D had lower LDH levels. Treatment with various immunochemotherapy regimens was started in 17 (53.1%) patients. No significant difference was identified between levels of vitamin D and response to treatment in patients with CLL.

Vitamin D and MM

In all 37 (18 male, 19 female) patients with newly diagnosed MM, suboptimal serum levels of vitamin D (<30 ng/mL) were observed. One patient had vitamin D insufficiency (serum levels between 20–30 ng/mL), while the remaining 36 patients had vitamin D deficiency (levels below 20 ng/mL). Severe vitamin D deficiency (<10 ng/mL) was observed in 81% of patients. These results are presented in Figure 4. No statistically significant difference was observed between serum vitamin D levels in men (6.2 ± 5.2 mg/mL) and women (6.7 ± 6.1 mg/mL). There was also no significant difference in vitamin D level associated with age. However, the mean vitamin D level in patients 69–78 years old (4.1 ± 3.4 mg/mL) was significantly lower than that in patients

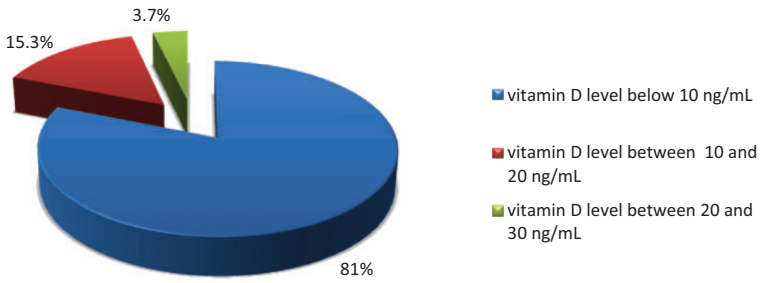

Figure 4. Distribution of patients with MM according to vitamin D level.

59 to 68 years old (9.4 ± 7.2 mg/mL). According to the Durie & Salmon staging system, 19 (51.4%) patients were in stage IIIB. Using the international staging system (ISS), 27 (73.0%) patients were in stage III. No statistically significant association was observed between serum vitamin D levels and clinical stage using either staging system. No statistically significant association was observed between vitamin D levels and response to treatment. No statistically significant association was observed between vitamin D levels and response to first-line therapy in patients with MM.

Discussion

Many data have been accumulated assessing the effects of vitamin D on cellular differentiation and proliferation, as well as angiogenesis and apoptosis.[10] Low serum levels of vitamin D were associated with a higher risk of developing NHLs.[11,12] Conversely, more frequent and longer sun exposure, which leads to higher serum levels of vitamin D, reduces the risk of developing NHLs.[13] van der Rhee et al.[14] stated that almost all epidemiological studies have documented an association between long-term sun exposure and reduced risks of colon, breast and prostate cancers as well as NHLs. In a prospective study of 155 patients with NHL-DLBCL, Hohaus et al.[15] observed vitamin D deficiency (<20 mg/mL) in 105 patients (67%), insufficiency (20–30 ng/mL) in 32 patients (21%) and normal levels (>30 ng/mL) in 18 patients (12%). The results of this study were similar to our own: we observed vitamin D insufficiency in 5.9% of patients, deficiency in 61.8% of patients and severe deficiency in 32.3% of patients with NHL-DLBCL. Molica et al. studied 130 patients with B-cell CLL and observed a deficiency of vitamin D in 82.3% of patients. Severe vitamin D deficiency (<10 ng/mL) was observed in 41 patients (31.5%), moderate deficiency (10–24 ng/mL) was observed in 66 patients (50.24%) and optimal vitamin D levels (25–80 ng/mL) were observed in 23 patients. No association was identified between vitamin D levels and season.[16] In our study, we observed vitamin D deficiency in 34.4% of patients, deficiency in 66.6% of patients, and severe deficiency in 28.1% of patients with CLL. In a study of 148 patients with newly diagnosed MM, vitamin D deficiency (<20 mg/mL) was observed in 24% of patients. According to the ISS, vitamin D deficiency was observed in 16% of patients at stage I, 20% of patients at stage II, and 37% of patients at stage III. Patients with low vitamin D levels had elevated C-reactive protein levels as well as high serum creatinine levels. In our study, no statistically significant associations were observed between levels of vitamin D and clinical stage, nor

between levels of vitamin D and those of C-reactive protein and creatinine. Previous studies found no correlation between vitamin D levels and myeloma bone disease, in agreement with our results.[17]

In conclusion, we observed low serum vitamin D levels in the majority of patients newly diagnosed with NHL-DLBCL, CLL and MM. Despite these unidirectional results, the inclusion of three cohorts of patients was an advantage, enabling comparisons between the three groups. It was also a disadvantage because of the different nature of these three diseases. Our study was limited in power because of the low number of patients included in each group and the lack of a control group of healthy individuals. Our results warrant further evaluation in other prospective studies, ideally using a large number of patients and a matched healthy control group. Because the prevalence of vitamin D deficiency appears to be high in different hematological malignancies, further studies evaluating the impact of maintaining adequate vitamin D levels on treatment and survival as week as prevention of secondary neoplasms is needed.

Declaration of conflicting interest

The authors declare that there is no conflict of interest.

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