ISSN (electrónico): 1699-5198 - ISSN (papel): 0212-1611 - CODEN NUHOEQ S.V.R. 318

ARÁN

Nutrición Hospitalaria

Trabajo Original

Otros

Vitamin D status and predictors of 25-hydroxyvitamin D levels in patients with heart failure living in a sunny region

Estado de la vitamina D y predictores de los niveles de 25-hidroxivitamina D en pacientes con insuficiencia cardíaca residentes en una región soleada

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Abstract

Aims: hypovitaminosis D has frequently been identified in patients with heart failure (HF). However, few studies have been conducted in regions with high solar incidence. Therefore, this study aimed to evaluate vitamin D status and predictors of 25-hydroxyvitamin D (25(OH)D) levels in patients with HF living in a sunny region (5 °- 6 °S).

Methods: this cross-sectional study enrolled 70 patients with HF. Biodemographic, clinical, biochemical, dietary, and sun exposure data were collected, and 25(OH)D levels were measured.

Results: the mean 25(OH)D level was 40.1 (12.4) ng/mL, and 24.3 % (95 % CI: 14.2-33.8) of patients with HF had hypovitaminosis D (25(OH)D < 30 ng/mL). Female patients ($p = 0.001$), those with ischemic etiology ($p = 0.03$) and those with high parathyroid hormone levels (> 67 pg/mL) ($p = 0.034$) were more likely to present hypovitaminosis D. Higher 25(OH)D levels were observed in men than in women ($\beta = 7.78$, $p = 0.005$) and in patients with HF in New York Heart Association (NHYA) functional class I when compared to those in class III/IV ($\beta = 8.23$, $p = 0.032$).

Conclusions: the majority of patients with HF had sufficient 25(OH)D levels. Sex and functional classification were identified as independent predictors of 25(OH)D levels. These results highlight the need for increased monitoring of vitamin D status among female patients with heart failure and those with more severe symptoms.

Keywords:

Heart failure. Cardiovascular disease. Vitamin D. Hypovitaminosis D. Nutritional status.

Received: 18/08/2020 • Accepted: 24/12/2020

Declaration of conflicts of interest: the authors declare no potential conflicts of interest concerning the research, authorship, and publication of this article.

Statement on funding sources: this work was supported by the Rio Grande do Norte Research Foundation (Fundação de Apoio à Pesquisa do Rio Grande do Norte – FAPERN; Brazil)/CAPES, Brazil under Grant 006/2014; and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001.

Statement of authorship: RCSDK, NRDL, RVZD and KCMSE conceived and designed the study; KCMSE was in charge of funding acquisition and project administration; RCSDK, FLA, NRDL and RVZD performed the collection of data. RCSDK and KCMS assembled, analyzed, and interpreted the data. RCSDK, FLA, NRDL, RVZD, SCVCL, LFCP and KCMSE drafted the article and revised it critically for intellectual content. All authors read and approved the final version of the article to be submitted.

Consent for publication: all authors agreed to the submission and approved the final version of the manuscript.

Acknowledgements: the authors thank the laboratory staff for performing the biochemical analyses; the students Ana Francisca Gomes, Eduardo da Silva, Giovanna Carvalho, Isabelli da Silva, Jainara Soares, and Larissa Silva for their assistance with data collection, and the Coordination for the Improvement of Higher Education Personnel (Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – CAPES) for providing a scholarship grant.

Dantas-Komatsu RCS, Freire FLA, Lira NRD, Diniz RVZ, Lima SCVC, Pedrosa LFC, Sena-Evangelista KCM. Vitamin D status and predictors of 25-hydroxyvitamin D levels in patients with heart failure living in a sunny region. Nutr Hosp 2021;38(2):349-357

DOI: http://dx.doi.org/10.20960/nh.03291

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Resumen

Objetivos: la hipovitaminosis D se ha identificado con frecuencia en pacientes con insuficiencia cardíaca (IC). Sin embargo, pocos estudios se han realizado en regiones con una alta exposición solar. Por lo tanto, este estudio tuvo como objetivo evaluar el estado de la vitamina D y los predictores de los niveles de 25-hidroxivitamina D (25(OH)D) en pacientes con IC que viven en una región soleada (5 °-6 °S).

Métodos: este estudio transversal incluyó a 70 pacientes con IC. Se recopilaron datos biodemográficos, clínicos, bioquímicos, dietéticos y de exposición solar, y se midieron los niveles de 25(OH)D.

Resultados: el nivel medio de 25(OH)D fue de 40,1 (12,4) ng/mL y el 24,3 % (IC 95 %: 14,2-33,8) de los pacientes con IC tenían hipovitaminosis D (25(OH)D < 30 ng/mL. Las pacientes mujeres (p = 0,001), aquellos con IC de etiología isquémica (p = 0,03) y aquellos otros pacientes con niveles altos de hormona paratiroidea (> 67 pg/mL) (p = 0,034) tenían más probabilidades de presentar hipovitaminosis D. Se observaron niveles más altos de 25(OH)D en los hombres que en las mujeres (β = 7,78, p = 0,005), y en los pacientes con IC de clase funcional I de la New York Heart Association (NHYA) que en los de clase III/IV (β = 8,23, p = 0,032).

Conclusiones: la mayoría de los pacientes con IC tenían niveles suficientes de 25(OH)D. El sexo y la clasificación funcional se identificaron como predictores independientes de los niveles de 25(OH)D. Estos resultados destacan la necesidad de un mayor control del estado de la vitamina D entre las mujeres con insuficiencia cardíaca y los pacientes con síntomas más graves.

Palabras clave:

Insuficiencia cardíaca. Enfermedad cardiovascular. Vitamina D. Hipovitaminisis D. Estado nutricional.

INTRODUCTION

Heart failure (HF) is a clinical syndrome characterized by structural and/or functional cardiac abnormalities that impair ventricular filling or blood ejection, resulting in reduced cardiac output and/or elevated intracardiac pressure at rest and during stress. Furthermore, HF involves complex metabolic, immunological, and neuroendocrine interactions that can negatively affect muscle, fat, and bone metabolism (1).

Vitamin D has been recognized for its key role as a mediator of calcium and phosphorus homeostasis in bones (2). Additionally, vitamin D is also involved in extra-skeletal tissues (3); in particular, the specific nuclear vitamin D receptor (VDR) and enzymes associated with vitamin D metabolism have been observed in tissues other than bones, including in the cardiovascular system (4). 1,25-dihydroxy-vitamin D [1,25$(OH)_2$D], the active vitamin D metabolite, exerts effects on the heart since it binds to the vitamin D receptor (VDR), and has potential modulatory effects on cardiac hypertrophy and failure (5); it also has a regulatory role in: a) the renin-angiotensin-aldosterone system; b) the remodelling and mediation of collagen deposition, fibrosis, and matrix metalloproteinase levels in the cardiac system; c) immediate and non-genomic effects on signal transduction mediators, and d) the modulation of the inflammatory process (3-5).

Low concentrations of vitamin D are frequently found in patients with HF, and they have been associated with increased plasma renin activity, inflammation profile, and high concentrations of natriuretic peptides and parathormone (PTH). These changes impair renal function, thereby worsening the functional capacity and severity of HF (6-9). In most cases, hypovitaminosis D is attributed to low dietary intake of vitamin D and reduced sun exposure, which itself is attributable to low tolerance for exercise (10,11). However, most studies evaluating vitamin D status were conducted in geographical regions with higher latitudes and lower solar incidence (7-9,12,13), and failed to consider factors directly related to vitamin D uptake, including sun exposure, skin type, and dietary intake (2). To address this gap, our study aimed to evaluate vitamin D status and predictors of 25(OH)D levels in patients with HF living in a sunny region of north-eastern Brazil.

MATERIAL AND METHODS

STUDY POPULATION AND ETHICAL ASPECTS

This analytical cross-sectional study included adult and elderly subjects of both sexes aged ≥ 18 years who were diagnosed with HF at Onofre Lopes University Hospital in Natal (5° 45' 54" S), Rio Grande do Norte, Brazil. Natal, a city in north-eastern Brazil, has a high solar radiation intensity, with an ultraviolet index > 7 year-round and levels > 10 between October and February (14)

Clinical diagnoses of HF were confirmed by a team of cardiologists, according to the Framingham and Boston criteria, as well as Doppler echocardiography (15). We excluded patients with a previous diagnosis of cancer, autoimmune disease and/or osteoporosis; haemodialysis or peritoneal dialysis; history of bariatric surgery; hepatic transaminase levels greater than three times the reference values; and patients who were pregnant or lactating. Patients who used calcium, phosphorus, or vitamin D supplements, glucocorticoids, and/or antiepileptics during the last 3 months were also excluded.

Patients were selected by convenience sampling from the outpatient care flow between January and December 2017. At data collection, 120 patients were enrolled, of whom 40 were excluded because they did not meet the previously defined criteria. Additionally, five of the remaining 80 recruited patients refused to participate. Four patients dropped out and one died during the study. Finally, data were collected from 70 participants, who were stratified into two groups: "hypovitaminosis D" (n = 17) and "sufficient vitamin D" (n = 53).

The research proposal was approved by the institutional review board (protocol CAAE 59827516.2.0.0.0.5292). All participants were provided information about the study and signed an informed consent form.

CLINICAL, BIODEMOGRAPHIC AND ANTHROPOMETRIC DATA

Clinical data regarding the etiology of HF, New York Heart Association (NYHA) functional classification, presence of comorbidities,

and left ventricular ejection fraction (LVEF) value as assessed by Doppler echocardiography were obtained from electronic medical records. Patients with a LVEF < 40 %, 40-49 %, or ≥ 50 % were classified as having HF with a reduced ejection fraction (HFrEF), a mid-range ejection fraction (HFmrEF), or a preserved ejection fraction (HFpEF), respectively (1).

Physical activity practice was evaluated as previously reported (16). Skin type was defined according to Fitzpatrick (17), who classified human skin color into six categories, ranging from type I (white) to type VI (black). Sun exposure was assessed using a score corresponding to the last 7-day period prior to blood collection. First, a daily score that considered the time of sun exposure and the amount of exposed skin was calculated. The weekly score was obtained from the sum of daily scores and ranged from 0 to 56 hours (18). Considering a half-life of 3 weeks for 25(OH)D (19), each participant was allocated to the season of the year that corresponded to the date 21 days before blood collection.

Weight and height measurements were obtained using a digital scale with a capacity of 200 kg and a precision of 0.1 kg (Leader Balanças®, São Paulo, Brazil, LD-1050 Line, Model P-200 C). Height was measured using a stadiometer attached to the scale. The body mass index (BMI) was used as anthropometric assessment. BMI classifications for adults and elderly individuals were performed according to the World Health Organization (20) and Lipschitz (21), respectively.

BIOCHEMICAL ANALYSIS

Blood samples were collected via venipuncture after a 12-hour overnight fast. Blood glucose, triglycerides, HDL-c, albumin, and total calcium levels were determined via colorimetric-enzymatic assays. Phosphorus, glutamic-oxaloacetic transaminase (GOT), and glutamic-pyruvic transaminase (GPT) levels were measured using enzymatic ultraviolet (UV) assays. Glycated hemoglobin (HbA1c) and high-sensitivity C-reactive protein (hs-CRP) levels were analyzed by immunoturbidimetry. Creatinine was measured by the kinetic method, and urea by the UV kinetic method. All analyses were performed using the Wiener Lab® test kit (Wiener Laboratory Group, Santa Fe, Argentina) and CMD800iX1 automated device (Diamond Diagnostics, Holliston, MA, USA). LDL-c levels were determined using Friedewald's formula (22). A hemoglobin analysis was performed using the cyanide-free colorimetric method and the Advia® 2120i Haematology System (Siemens Healthcare, Erlangen, Germany). Serum intact PTH (iPTH) levels were determined by chemiluminescence using the commercially available Unicel® DxI800 immunoassay system (Beckman Coulter, Brea, CA, USA).

Serum 25(OH)D levels were evaluated using the COBAS 6000 Modular Equipment automated system (Roche®, Basel, Switzerland) and the Elecsys® Vitamin D total electrochemiluminescence immunoassay kit (Roche®, Basel, Switzerland). A cut-off point of 25(OH)D ≥ 30 ng/mL was used to define vitamin D sufficiency. Vitamin D deficiency was defined as serum 25(OH)D < 20 ng/ml (19). For this study, patients with 25(OH)D < 30 ng/mL were allocated to the hypovitaminosis D group (23). iPTH levels were also categorized; here, a level > 67 pg/mL was considered high. The estimated glomerular filtration rate (eGFR) was calculated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation, and a reduced eGFR was defined as a CKD-EPI result of < 60 mL/min/1.73 m^2 (1).

DIETARY INTAKE ASSESSMENT

The dietary intakes of vitamin D, calcium, and phosphorus were determined in triplicate by trained researchers using the 24-hour food recall method. A dietary analysis was performed using Virtual Nutri Plus® 2.0 (Keeple Company, São Paulo, Brazil). The Multiple Source Method was used to estimate the habitual intakes of nutrients and remove intrapersonal variability.

Subsequently, nutrients were adjusted by energy levels according to the method proposed by Willett and Stampfer (24). The prevalence of inadequate vitamin D intake was estimated using the equation below (25), with reference to the Estimated Average Requirement (EAR) (26).

$$z = (\text{mean of intake})/\text{standard deviation}$$

STATISTICAL ANALYSIS

Continuous variables are expressed as means (standard deviation) for normally distributed data, and by medians (quartiles 1-3) for asymmetrically distributed data. Categorical variables are expressed as absolute frequencies (percentages). The Shapiro–Wilk test was used to evaluate the distributions of data.

Data were missing for the following variables: sun exposure, total calcium, total cholesterol, HDL-c, LDL-c, fasting glucose, hemoglobin, HbA1c, iPTH, triglycerides, urea, dietary vitamin D, dietary calcium, and dietary phosphorus. In such cases, a simple imputation method was applied to replace the missing value with the central trend value of the variable. Imputation could not be performed for albumin, phosphorus, and hs-CRP levels because the amount of missing data exceeded the maximum allowable level.

Student's t-test and the Mann–Whitney test were used for comparisons of variables between groups according to 25(OH)D cut-off points as appropriate. The chi-square test was used to evaluate associations between categorical variables. Correlations between 25(OH)D levels and continuous variables were evaluated using Pearson's coefficient (r) for normally distributed data, and Spearman's coefficient for abnormally distributed data. The generalized linear regression model was adjusted for the variable 25(OH)D according to the stepwise method for the selection of model variables. We considered the independent variables to be clinically relevant for predicting 25(OH)D. The variables whose p-value was less than 0.10 in the bivariate analysis were included in the model. The criteria used to assess the permanence of the independent variable in the model were: 1) absence of

multicollinearity; 2) the Wald chi-squared test, and 3) the ability to improve the quality of the model using the Akaike information criterion (AIC). The initial predictors were sex, etiology, HF functional class, parathormone, and TGP (AIC = 572.69). However, after adjustment criteria were applied, the model selected to predict 25(OH)D levels was composed of sex, parathormone, and HF functional class (AIC = 565.24). For all analyses, a p-value < 0.05 was considered statistically significant. The software package SPSS Statistics® version 22.0 was used for data storage and analysis.

RESULTS

Most of the participants were male (64.3 %) with a mean age of 53(15) years, and 57.1 % of them were overweight. Furthermore, 60 % of the participants had HFrEF, and most met the criteria for NYHA functional class I. Female patients and those with an ischemic etiology were more likely to present with hypovitaminosis D, as well as patients who did not use angiotensin II receptor antagonists or angiotensin-converting enzyme inhibitors (ARA/ACEI), and those who did use antiplatelet medications (Table I).

Table I. Biodemographic, anthropometric, and clinical characteristics of patients with heart failure according to 25(OH)D levels

Variables	Total n = 70	25(OH)D		p-value
		Hypovitaminosis D < 30 ng/mL n = 17	Sufficient ≥ 30 ng/mL n = 53	
Age (years)[a]	53 (15)	58 (15)	52 (15)	0.119
Sex[b]				
Female	25 (35.7)	12 (70.6)	13 (24.5)	0.001*
Male	45 (64.3)	5 (29.4)	40 (75.5)	
BMI (kg/m²)[a]	26.63 (5.13)	26.29 (5.06)	26.73 (5.19)	0.762
Malnutrition	6 (8.6)	1 (1.4)	5 (7.1)	
Eutrophy	24 (34.3)	8 (11.4)	16 (22.9)	0.437
Overweight	40 (57.1)	8 (11.4)	32 (45.8)	
Skin type[b]				
II	7 (10)	2 (11.8)	5 (9.4)	0.611
III	26 (37.1)	7 (41.2)	19 (35.8)	
IV	15 (21.4)	5 (29.4)	10 (18.9)	
V	18 (25.7)	3 (17.6)	15 (28.3)	
VI	4 (5.7)	0 (0.0)	4 (7.5)	
Seasons[b]				
Summer	13 (18.6)	1 (5.9)	12 (22.6)	0.110
Autumn	25 (35.7)	4 (23.5)	21 (39.6)	
Winter	23 (32.9)	9 (52.9)	14 (26.4)	
Spring	9 (12.9)	3 (17.6)	6 (11.3)	
Solar exposure score[a]	15 (6-28)	10.0 (4.0-23.5)	16.0 (6.5-28.0)	0.232
Sunscreen	12 (17,1)	3 (17,6)	9 (17,0)	0.949
Exercise time (min/week)[c]	37.5 (0.0-150.0)	0.0 (0.0-175.0)	75.0 (0.0-150,0)	0.389
Etiology[b]				
Ischemic	29 (41.4)	11 (64.7)	18 (34.0)	0.030*†
Non-ischemic	30 (42.9)	4 (23.5)	26 (49.1)	
Undefined	11 (15.7)	2 (11.8)	9 (17.0)	
LVEF (%)[c]	36 (26-50)	41 (28-54)	34.5 (26-47)	0.255

(Continuation in the next page)

Table I (Cont.). Biodemographic, anthropometric, and clinical characteristics of patients with heart failure according to 25(OH)D levels

Variables	Total n = 70	25(OH)D		p-value
		Hypovitaminosis D < 30 ng/mL n = 17	Sufficient ≥ 30 ng/mL n = 53	
HF definition[b]				
HFrEF	42 (60.0)	8 (47.1)	34 (64.2)	
HFmrEF	10 (14.3)	3 (17.6)	7 (13.2)	0.448
HFpEF	18(25.7)	6 (35.3)	12 (22.6)	
NYHA functional classification[b]				
I	46 (65.7)	8 (47.1)	38 (71.7)	
II	14 (20.0)	4 (23.5)	10 (18.9)	0.085
III/IV	10(14.3)	5 (29.4)	5 (9.4)	
Comorbidity[b]				
Arterial hypertension	34 (48.6)	8 (47.1)	26 (49.1)	0.886
Diabetes *mellitus*	19 (27.1)	6 (35.3)	13 (24.5)	0.385
eGFR < 60 mL/min/1.73 m^2	21 (30)	5 (29.4)	16 (30.2)	0.951
Medication[b]				
ACEI/ARA	66 (94.3)	14 (82.4)	52 (98.1)	0.015*
Diuretics	57 (81.4)	16 (94.1)	41 (77.4)	0.122
β-blockers	65 (92.9)	15 (88.2)	50 (94.3)	0.395
Vasodilators	17 (24.3)	3 (17.6)	14 (26.4)	0.463
Digoxin	15 (21.4)	3 (17.6)	12 (22.6)	0.662
Antiplatelet agents	32 (45.7)	13 (76.5)	19 (35.8)	0.003*

*[a]Data are presented as means (standard deviation) with p-values determined using a t test; [b]Data are presented as n (%) with p-values determined using the χ^2 test; [c]Data are presented as medians (quartiles 1-3) with p-values determined using the Mann–Whitney test. *Statistically significant comparison between the groups (p < 0.05). [†]p-value for comparison of ischemic vs. non-ischemic etiology; 25(OH)D: 25-hydroxyvitamin D; BMI: body mass index; eGFR: estimated glomerular filtration rate; LVEF: left ventricular ejection fraction; HFpEF: heart failure with preserved ejection fraction; HFmrEF: heart failure with mid-range ejection fraction; HFrEF: heart failure with reduced ejection fraction; NYHA: New York Heart Association; ACEI: angiotensin-converting enzyme inhibitor; ARA: angiotensin II receptor antagonist.*

Overall, the mean serum 25(OH)D level was 40.1 (12.4) ng/mL, and the frequency of hypovitaminosis D was 24.3 % (95 % CI: 14.2-33.8). Vitamin D deficiency was recorded in 7.1 % (95 % CI: 1.1-13.2) of participants with HF. In the total group, 19 patients (27.1 %) had a high iPTH level (> 67 pg/mL), and those individuals were more likely to present hypovitaminosis D ($p = 0.034$). The prevalence of inadequate vitamin D intake was 100 %, with no difference in mean intake between the groups ($p \geq 0.05$). No significant inter-group differences were observed in other analyses of biochemical and dietary variables (all $p \geq 0.05$) (Table II).

Significant positive correlations were observed between 25(OH)D levels and albumin ($r = 0.365$, $p = 0.015$), total calcium ($r = 0.266$, $p = 0.026$), hemoglobin ($r = 0.249$, $p = 0.037$), and GPT ($r = 0.366$, $p = 0.002$) levels. Furthermore, a significant, negative correlation was observed between 25(OH)D and iPTH levels ($r = -0.255$; $p = 0.033$) (Table III).

In a regression model adjusted for 25(OH)D, the NYHA functional classification, iPTH category, and sex were identified as variables with a better adjustment quality. This analysis indicated that men with HF had significantly higher 25(OH)D levels as compared to women ($\beta = 7.78$, $p = 0.005$). Additionally, participants in the NYHA class I had significantly higher 25(OH)D levels when compared to those in NYHA class III/IV ($\beta = 8.23$, $p = 0.032$). Although a high iPTH level (> 67 pg/mL) was correlated inversely with 25(OH)D level ($\beta = -5.52$, $p = 0.062$), this association was not statistically significant (Fig. 1, Table IV).

DISCUSSION

This is the first study to assess vitamin D status in patients with HF living in a region of north-eastern Brazil with high solar radiation intensity. Most patients with HF were clinically stable (NYHA I) and had a sufficient 25(OH)D status. However, we observed significant associations of 25(OH)D levels with clinical and biochemical parameters. We identified sex and functional classifi-

Table II. Biochemical and dietary data of patients with heart failure according to 25(OH)D levels

Variables	Total n = 70	25OHD		p-value
		Hypovitaminosis D < 30 ng/mL n = 17	Sufficient ≥ 30 ng/mL n = 53	
25(OH)D (ng/mL)[a]	40.1 (12.4)	22.85 (4.1)	45.62 (8.5)	< 0.001*
iPTH (pg/mL)[b]	49.15 (35.9-75.2)	66.5 (44.0-112.1)	46.9 (34.95-64.9)	0.020*
iPTH > 67 pg/mL[c]	19 (27.1)	8 (47.1)	11 (20.8)	0.034*
iPTH ≤ 67 pg/mL[c]	51 (72.9)	9 (52.9)	42 (79.2)	
Total calcium (mg/dL)[a]	10.0 (0.6)	9,8 (0.8)	10.1 (0.6)	0.175
Phosphorus (mg/dL)[a,†]	3.8 (0.6)	3.6 (0.5)	3.84 (0.6)	0.208
Hemoglobin (g/dL)[a]	13.2 (1.7)	12.8 (2.0)	13.4 (1.5)	0.258
Fasting glycemia (mg/dL)[b]	103.0 (95.8-120.3)	109 (94-155)	103 (96.0-117.5)	0.279
HbA1c (%)[b]	6.3 (6.0-6.7)	6.3 (5.9-7.35)	6.3 (6.0-6.6)	0.721
Total cholesterol (mg/dL)[b]	160.5 (1323-182.3)	165.0 (125.5-191.5)	159 (131.5-180.5)	0.529
HDL-c (mg/dL)[a]	35.4 (8.0)	35.7 (7.8)	35.3 (8.1)	0.860
LDL-c (mg/dL)[b]	89.0 (70.8-110.8)	94 (74-113.5)	89 (69.5-1100)	0.661
Triglycerides (mg/dL)[b]	119.0 (76.5-185.5)	129.0 (53.0-217.0)	119.0 (77.0-163.5)	0.701
GOT (U/L)[b]	22.5 (18.0-29.0)	22.00 (17.5-33,0)	23.00 (19.0-29.0)	0.995
GPT (U/L)[b]	23.0 (18.0-29.5)	21.0 (15.0-25.5)	23.0 (19.0-31.5)	0.236
Albumin (g/dL)[b,†]	4.4 (4.2-4.7)	4.4 (3.6-4.5)	4.4 (4.3-4.75)	0.104
Creatinine (mg/dL)[b]	1.1 (0.9-1.3)	1.0 (09-1.3)	1.1 (0.9-1.3)	0.699
Urea (mg/dL)[b]	38.0 (31.5-55.0)	38.0 (33-56.5)	38.00 (30.0-54.5)	0.722
hs-CRP (mg/L)[b,†]	1.0 (0.3-6.7)	2.9 (0.5-13.9)	0.8 (0.3-5.6)	0.189
eGFR (mL/min/1.73 m^2)[a]	73.8 (24.9)	65.5 (21.3)	76.5 (25.5)	0.115
Vitamin D intake (µg/d)[a]	2.6 (1.2)	2.7 (1.2)	2.6 (1.2)	0.725
Calcium intake (mg/d)[b]	354.7 (313.5-487.1)	383.2 (325.7-483.0)	349.19 (308.9-487.5)	0.493
Phosphorus intake (mg/d)[a]	766.7 (147.9)	766.05 (92.9)	766.96 (162.3)	0.983

*[a]Data are presented as means (standard deviation) with p-value determined using the t-test; [b]Data are presented as medians (quartiles 1-3) with p-values determined using the Mann-Whitney test; [c]Data are presented as n (%) with p-values determined using the χ^2 test; [†]Phosphorus, n = 54 (12/42); albumin, n = 44 (15/29); hs-CRP, n = 58 (15/43); *Statistically significant comparison between the groups (p < 0.05); 25(OH)D: 25-hydroxyvitamin D; iPTH: intact parathyroid hormone; HbA1Ac: glycated hemoglobin; GOT: glutamic-oxaloacetic transaminase; GPT: glutamic-pyruvic transaminase; hs-CRP: high-sensitivity C-reactive protein; eGFR: estimated glomerular filtration rate.*

cation as independent predictors of 25(OH)D levels; specifically, we observed lower vitamin D levels in female participants and in those with worse functional NYHA classification.

Our findings regarding the 25(OH)D status are in contrast with those studies of HF patients from various populations. Generally, studies conducted in higher-latitude regions reported a high prevalence of vitamin D deficiency (7-9,12). Additionally, a previous study in a sample of patients with HF living in Rio de Janeiro (22 °S) in Brazil reported that 47 % of patients had hypovitaminosis D (< 30 ng/mL) (27); another study in Porto Alegre (30 °S) reported a rate of 70.5 % (13). Although these studies were also conducted in Brazil, the authors used different methods of analysis for the quantification of 25(OH)D; for example, radioimmunoassay (13) and liquid chromatography (27), thus limiting the comparability of studies. However, the Elecsys Vitamin D Total assay used in our study demonstrated a good overall performance, and was very suitable for automated measurement of 25(OH)D (28).

In addition, these Brazilian studies carried out in patients with HF were conducted in southeastern and southern regions of Brazil, which are different from the north-eastern region where this study was performed. The angle of solar radiation increases with increasing latitude; therefore, in regions at latitudes distant from the equator, UV-B rays are absorbed to a greater extent by the ozone layer and little radiation reaches the earth's surface, reducing the efficiency of vitamin D production (2). Likely, this factor best explains the relatively lower frequency of hypovitaminosis D

Table III. Correlations between 25(OH)D concentrations and clinical parameters in all patients with heart failure

Variables	25(OH)D	
	R	p-value
Age[a]	-0.108	0.372
BMI[a]	-0.004	0.972
LVEF[b]	-0.125	0.302
Total calcium[a]	0.266	0.026*
iPTH[b]	-0.255	0.033*
Phosphorus[a,†]	0.158	0.253
Hemoglobin[a]	0.249	0.037*
Albumin[b,†]	0.365	0.015*
GOT[b]	-0.232	0.054
GPT[b]	0.366	0.002*
hs-CRP[b,†]	-0.131	0.328
eGFR[a]	0.157	0.195
Solar exposure score[b]	0.174	0.150
Vitamin D intake[a]	0.088	0.466
Calcium intake[b]	0.046	0.704
Phosphorus intake[a]	0.09	0.436

*[a]Pearson's correlation; [b]Spearman's correlation; *Statistically significant at a level of p < 0.05; †Phosphorus, n = 54; albumin, n = 44; hs-CRP, n = 58; 25(OH)D: 25-hydroxyvitamin D; BMI: body mass index; LVEF: left ventricular ejection fraction; iPTH: intact parathyroid hormone; GOT: glutamic-oxaloacetic transaminase; GPT: glutamic-pyruvic transaminase; hs-CRP: high-sensitivity C-reactive protein; eGFR: estimated glomerular filtration rate.*

Table IV. Stepwise forward regression analysis of 25(OH)D predictors

Variable	β	Standard error	p-value
Intercept	30.55	3.90	< 0.000
Sex			
Male	7.78	2.75	0.005*
Female	0		
PTH			
> 67 pg/mL	-5.52	2.96	0.062
≤ 67 pg/mL	0		
NYHA functional classification			
I	8.23	3.83	0.032*
II	3.09	4.54	0.495
III/IV	0		

**Statistically significant at a level of p < 0.05; 25(OH)D: 25-hydroxyvitamin D; NYHA: New York Heart Association.*

and vitamin D deficiency observed in our study, considering that Natal has a latitude between 5 ° and 6 ° south of the equator, resulting in high solar radiation levels throughout the year (14).

Although the associations observed between sun exposure, dietary vitamin D intake, and 25(OH)D levels were not significant in this study, vitamin D status has been discussed not only by dietary intake or endogenous production, but also by the body's ability to store and release the vitamin (29). Lifelong exposure to a high ultraviolet index may provide adequate endogenous vitamin D production and storage. The mechanisms underlying 25(OH)D storage and bioavailability are still unclear, but cell culture studies suggest that PTH (30) and 1,25-dihydroxyvitamin D [$1,25(OH)_2D$] (31) regulate the uptake and release of vitamin D by skeletal and adipose muscle cells. Thus, it is suggested that the high frequency of vitamin D sufficiency observed in our study possibly reflected the bioavailability of 25(OH)D stored in the patients' tissues.

We observed a higher likelihood of hypovitaminosis D among patients with HF of ischemic etiology as compared to those with non-ischemic etiology. Vitamin D deficiency appears to contribute to an imbalance in vascular homeostasis and compromises arterial functioning, affecting the progression of atherosclerosis (32).

The observation that women displayed lower 25(OH)D levels and faced a greater risk of hypovitaminosis D relative to men should be emphasised; notably, these findings agree with the results of other studies (7,33). Sex steroid hormones regulate the synthesis of hepatic vitamin D-binding protein, the major vitamin D carrier protein. In postmenopausal women reduced estrogen levels appear to imply a lower total vitamin D level (34). We note that 64 % of the women in our study were menopausal. In addition, fat mass significantly increased between premenopausal and postmenopausal women when evaluated by BMI. Body mass can reduce proportionally circulating vitamin D levels (35). Although there was no significant correlation between BMI and 25(OH)D in our study, the body composition changes of menopause may explain the identification of sex as an independent predictor of 25(OH)D levels.

We identified the variable of functional classification as a predictor of vitamin D status. Previous reports regarding the associations of low 25(OH)D levels with highest symptom severity, determined by the functional classification, have been conflicting (7,11,36,37). However, this relationship is supported by evidence from other parameters that indicate clinical severity, such as associations between low 25(OH)D levels and elevated natriuretic peptide levels (7,36), reduced functional capacity (9), and the occurrence of poor outcomes in HF (6,7,37).

A previous meta-analysis confirmed an association of the most serious functional classification with increased bone loss, and an imbalance in calcium and phosphorus homeostasis, as well as in the hormones involved in this balance, have been identified

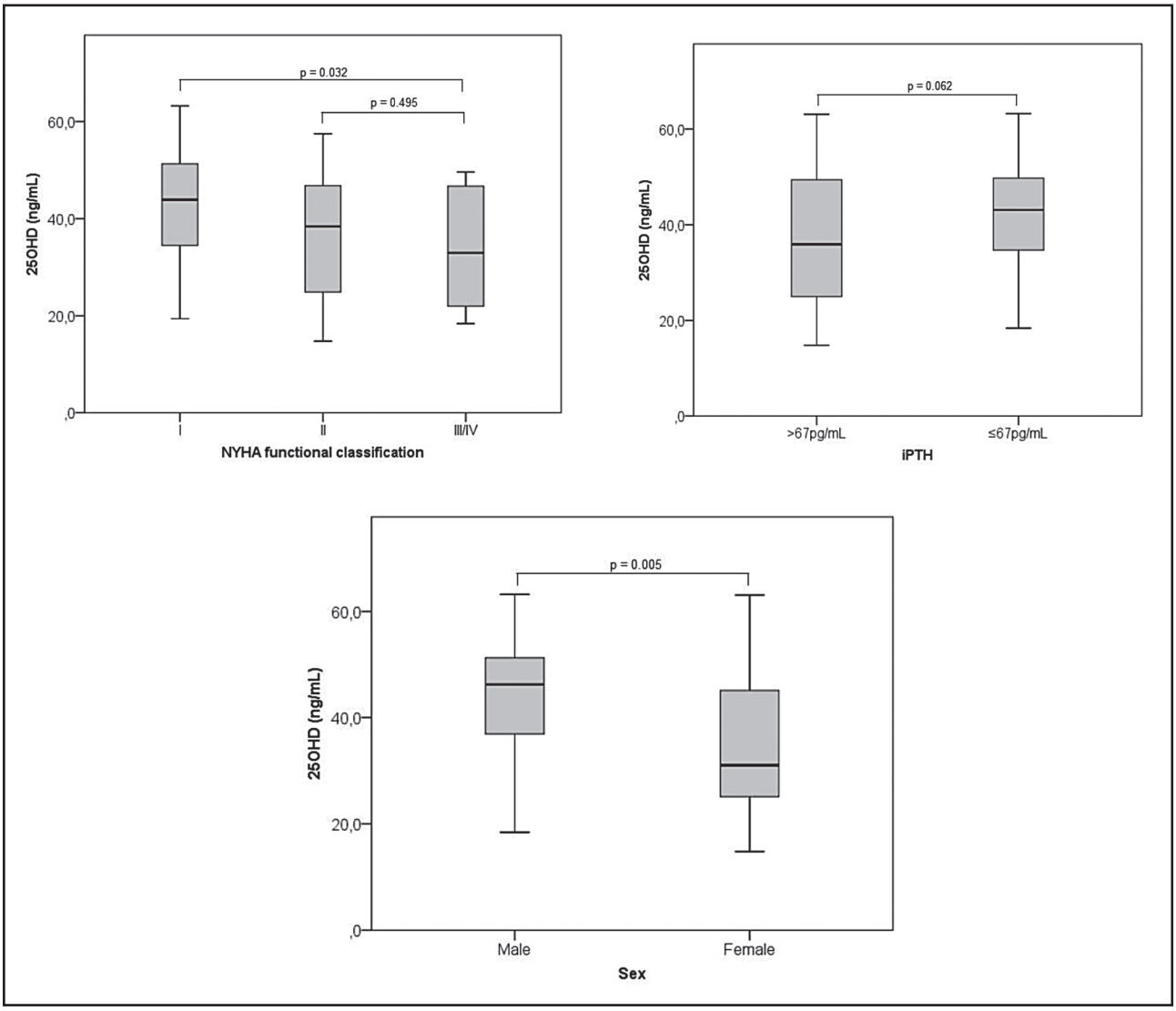

Figure 1.

Differences in 25(OH)D levels by NYHA functional class, iPTH category, and sex. 25(OH)D: 25-hydroxyvitamin D; NYHA: New York Heart Association; iPTH: intact parathyroid hormone.

as predictors of clinical outcome in end-stage HF patients (38). Given the role of vitamin D in maintaining the balance of calcium and phosphorus, patients with more clinically severe HF may have an increased requirement for vitamin D. Furthermore, we confirmed the associations of lower 25(OH)D levels with lower serum calcium and higher PTH levels. Similar findings have been previously reported (8,10,39), including an observation that higher HF-related mortality was associated with elevated PTH levels and hypovitaminosis D (10).

Our study had several limitations. First, the cross-sectional design of the study prevented us from establishing a causal relationship between hypovitaminosis D and HF. Second, although the data collection had taken place in a heart failure center, we obtained a small sample. Third, the biochemical assay used to evaluate 25(OH)D may have cross-reacted with other vitamin D metabolites (40), and the sun exposure instrument can only evaluate sun exposure in the 7 previous days.

CONCLUSION

In conclusion, a majority of patients with HF residing in a sunny region had sufficient 25(OH)D levels. Sex and functional classification were identified as independent predictors of 25(OH)D levels, with low 25(OH)D levels among women and patients with HF in NYHA class III/IV. Our results reinforce the importance of monitoring female patients with HF, and those with worse symptomatic severity to prevent poor outcomes related to hypovitaminosis D.

REFERENCES

1. Ponikowski P, Voors AA, Anker SD, Bueno H, Cleland JGF, Coats AJS, et al. 2016 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure. Eur Heart J 2016;37:2129-200. DOI: 10.1093/eurheartj/ehw128
2. Holick M. F. Vitamin D: Evolutionary, physiological and health perspectives. Curr Drug Targets 2011;12:4-18. DOI: 10.2174/138945011793591635
3. Wimalawansa SJ. Non-musculoskeletal benefits of vitamin D. J Steroid Biochem Mol Biol 2018;175:60-81. DOI: 1016/j.jsbmb.2016.09.016
4. Chen S, Glenn DJ, Ni W, Grigsby CL, Olsen K, Nishimoto M, et al. Expression of the Vitamin D receptor is increased in the hypertrophic heart. Hypertension 2008;52:1106-12. DOI: 10.1161/HYPERTENSIONAHA.108.119602
5. Saponaro, F., Marcocci, C. & Zucchi, R. Vitamin D status and cardiovascular outcome. J Endocrinol Invest 2019;42:1285-90. DOI: 10.1007/s40618-019-01057-y
6. Zittermann A, Schleithoff SS, Götting C, Dronow O, Fuchs U, Kuhn J, et al. Poor outcome in end-stage heart failure patients with low circulating calcitriol levels. Eur J Heart Fail 2008;10(3):321-7.
7. Liu LCY, Voors AA, van Veldhuisen DJ, van der Veer E, Belonje AM, Szymanski MK, et al. Vitamin D status and outcomes in heart failure patients. Eur J Heart Fail 2011;13:619-25.
8. Schierbeck LL, Jensen TS, Bang U, Jensen G, Køber L, Jensen J-EB. Parathyroid hormone and vitamin D--markers for cardiovascular and all cause mortality in heart failure. Eur J Heart Fail 2011;13:626-32.
9. Saponaro F, Marcocci C, Zucchi R, Prontera C, Clerico A, Scalese M, et al. Hypovitaminosis D in patients with heart failure: effects on functional capacity and patients' survival. Endocrine 2017;58:574-81.
10. Zittermann A, Schleithoff SS, Tenderich G, Berthold HK, Körfer R, Stehle P. Low vitamin D status: a contributing factor in the pathogenesis of congestive heart failure? J Am Coll Cardiol 2003;41:105-12. DOI: 10.1016/S0735-1097(02)02624-4
11. Belen E, Tipi FF, Aykan AC, Findikçioğlu U, Karakuş G, Yeşil A, et al. Clinical staging in chronic heart failure associated with low vitamin D and elevated parathormone levels. Acta Cardiol 2014;69:665-71. DOI: 10.2143/AC.69.6.1000009
12. Kim DH, Sabour S, Sagar UN, Adams S, Whellan DJ. Prevalence of hypovitaminosis D in cardiovascular diseases (from the National Health and Nutrition Examination Survey 2001 to 2004). Am J Cardiol 2008;102:1540-4. DOI: 10.1016/j.amjcard.2008.06.067
13. Lopes MR, Ribeiro PAB, Ledur P, Souza GC, Clausell N, Schaan BD. Vitamin D insufficiency is associated with lower physical function in patients with heart failure and diabetes. J Diabetes Res 2014;2014:1-9. DOI: 10.1155/2014/320930
14. Instituto Nacional de Pesquisas Espaciais (INPE). Radiação UV Natal. Natal (RN/Brazil): Laboratório de variáveis ambientais tropicais, Ministério da Ciência, Tecnologia e Inovação; 2012 [Accessed Oct 25, 2018]. Available at: http://www.crn.inpe.br/lavat/index.php?id=radiacaouvnt/
15. Bocchi E, Marcondes-Braga F, Bacal F, Ferraz A, Albuquerque D, Rodrigues D. Atualização da Diretriz Brasileira de Insuficiência Cardíaca Crônica - 2012. Arq Bras Cardiol 2012;98:1-33.
16. Brasil. Ministério da Saúde. Vigitel Brasil 2016 Saúde Suplementar : vigilância de fatores de risco e proteção para doenças crônicas por inquérito telefônico. Brasília: Ministério da Saúde; 2017. p. 157 [Accessed Jan 15, 2018]. Available at: https://www.ans.gov.br/images/Vigitel_Saude_Suplementar.pdf.
17. Fitzpatrick TB. The validity and practicality of sun-reactive skin types I through VI. Arch Dermatol 1988;124:869-71. DOI: 10.1001/archderm.1988.01670060015008
18. Hanwell HEC, Vieth R, Cole DEC, Scillitani A, Modoni S, Frusciante V, et al. Sun exposure questionnaire predicts circulating 25-hydroxyvitamin D concentrations in Caucasian hospital workers in southern Italy. J Steroid Biochem Mol Biol 2010;121:334-7. DOI: 10.1016/j.jsbmb.2010.03.023
19. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, et al. Evaluation, treatment, and prevention of vitamin D deficiency: An endocrine society clinical practice guideline. J Clin Endocrinol Metab 2011;96:1911-30. DOI: 10.1210/jc.2011-0385
20. World Health Organization. Obesity : preventing and managing the global epidemic : report of a WHO consultation. Geneva; 2000.
21. Lipschitz DA. Screening for nutritional status in the elderly. Prim Care 1994;21:55-67.
22. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 1972;18:499-502. DOI: 10.1177/107424840501000106
23. Maeda SS, Borba VZ, Camargo MB, Silva DM, Borges JL, Bandeira F, et al. Brazilian Society of Endocrinology and Metabology (SBEM). Recommendations of the Brazilian Society of Endocrinology and Metabology (SBEM) for the diagnosis and treatment of hypovitaminosis D. Arq Bras Endocrinol Metabol 2014;58:411-33.
24. Willett W, Stampfer MJ. Total energy intake: implications for epidemiologic analyses. Am J Epidemiol 1986;124:17-27. DOI: 10.1093/oxfordjournals.aje.a114366
25. Institute of Medicine. Dietary Reference Intakes: Applications in Dietary Assessment. Washington, DC: The National Academies Press; 2000. p. 306. DOI: 10.17226/9956
26. Institute of Medicine. Dietary Reference Intakes for Calcium and Vitamin D. Washington, DC: The National Academies Press; 2011. p. 1132. DOI: 10.17226/13050
27. Jorge AJL, Rosa MLG, Freire MDC, Correia DM da S, Fernandes LCM, Ribeiro ML, et al. Vitamin D Deficiency among Patients with Suspected Heart Failure with Normal Ejection Fraction. Rev Bras Cardiol 2013;26:253-8.
28. Wielders JP, Carter GF, Eberl H, Morris G, Roth HJ, Vogl C. Automated Competitive Protein-Binding Assay for Total 25-OH Vitamin D, Multicenter Evaluation and Practical Performance. J Clin Lab Anal 2015;29(6):451-61. DOI: 10.1002/jcla.21793
29. Abboud M, Puglisi DA, Davies BN, Rybchyn M, Whitehead NP, Brock KE, et al. Evidence for a specific uptake and retention mechanism for 25-hydroxyvitamin D (25OHD) in skeletal muscle cells. Endocrinology 2013;154:3022-30. DOI: 10.1210/en.2012-2245
30. Abboud M, Rybchyn MS, Liu J, Ning Y, Gordon-Thomson C, Brennan-Speranza TC, et al. The effect of parathyroid hormone on the uptake and retention of 25-hydroxyvitamin D in skeletal muscle cells. J Steroid Biochem Mol Biol 2017;173:173-9. DOI: 10.1016/j.jsbmb.2017.01.001
31. Abboud M, Rybchyn MS, Ning YJ, Brennan-Speranza TC, Girgis CM, Gunton JE, et al. 1,25-Dihydroxycholecalciferol (calcitriol) modifies uptake and release of 25-hydroxycholecalciferol in skeletal muscle cells in culture. J Steroid Biochem Mol Biol 2017;177:109-15. DOI: 10.1016/j.jsbmb.2017.10.018
32. Huang J, Wang Z, Hu Z, Jiang W, Li B. Association between blood vitamin D and myocardial infarction: A meta-analysis including observational studies. Clin Chim Acta 2017;471:270-5. DOI: 10.1016/j.cca.2017.06.018
33. Costanzo S, De Curtis A, Di Castelnuovo A, Persichillo M, Bonaccio M, Pounis G, et al. Serum vitamin D deficiency and risk of hospitalization for heart failure: Prospective results from the Moli-sani study. Nutr Metab Cardiovasc Dis 2018;28:298-307. DOI: 10.1016/j.numecd.2017.11.008
34. Tsuprykov O, Chen X, Hocher C, Hocher B. Why should we measure free 25(OH) vitamin D? J Steroid Biochem Mol Biol 2018;180:87-104. DOI: 10.1016/j.jsbmb.2017.11.014
35. Delle Monache S, Di Fulvio P, Iannetti E, Valerii L, Capone L, Nespoli MG, et al. Body mass index represents a good predictor of vitamin D status in women independently from age. Clin Nutr 2019;38(2):829-34. DOI: 10.1016/j.clnu.2018.02.024
36. Pilz S, März W, Wellnitz B, Seelhorst U, Fahrleitner-Pammer A, Dimai HP, et al. Association of vitamin D deficiency with heart failure and sudden cardiac death in a large cross-sectional study of patients referred for coronary angiography. J Clin Endocrinol Metab 2008;93:3927-35. DOI: 10.1210/jc.2008-0784
37. Belen E, Sungur A, Sungur MA. Vitamin D levels predict hospitalization and mortality in patients with heart failure. Scand Cardiovasc J 2015;50:17-22. DOI: 10.3109/14017431.2015.1098725
38. Xing W, Xiaoling L, Wenyan G, Wang J, Yang Z, Wang S, et al. Bone mineral density in patients with chronic heart failure: a meta-analysis. J Pakistan Med Assoc 2018;13:343-53. DOI: 10.2147/CIA.S154356
39. Gotsman I, Shauer A, Zwas DR, Hellman Y, Keren A, Lotan C, et al. Vitamin D deficiency is a predictor of reduced survival in patients with heart failure; Vitamin D supplementation improves outcome. Eur J Heart Fail 2012;14:357-66. DOI: 10.1093/eurjhf/hfr175
40. Kocak FE, Ozturk B, Isiklar OO, Genc O, Unlu A, Altuntas I. A comparison between two different automated total 25-hydroxyvitamin D immunoassay methods using liquid chromatography-tandem mass spectrometry. Biochem Medica 2015;25:430-8. DOI: 10.11613/BM.2015.044