



Prevalence of Vitamin D Deficiency and its Association with Disease Severity in Children Hospitalized with Acute Respiratory Infections

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ABSTRACT: Background: Vitamin D deficiency (VDD) is highly prevalent in pediatric patients, and it has been implicated in the pathogenesis and clinical severity of acute respiratory infections (ARI). Objectives: To evaluate the prevalence of VDD and its association with ARI disease severity and clinical outcomes in hospitalized pediatric patients. Methods: A 4-year prospective case-control study, conducted between 2021 and 2024, in two Romanian pediatric hospitals, included 400 inpatients, aged 0-18 years, admitted for ARI. Patients were stratified into cases with a low level of vitamin D (n=200) and controls with a normal vitamin D (n=200). Clinical severity of ARI, inflammatory biomarkers, presence of fever, need for supplemental oxygen, and length of hospital stay were recorded and analyzed in both cases and controls. Results: Low serum vitamin D levels were significantly associated with severe ARI (26.5% vs. 5.5%), fever at admission (61.0% vs. 32.0%), and increased requirement for oxygen therapy (69.5% vs. 21.0%). Cases also exhibited higher C-reactive protein levels (3.50 vs. 1.64mg/L), higher clinical severity scores (3.77 vs. 1.62), and longer hospital length of stay (4.68 vs. 2.89 days). Conclusions: Vitamin D deficiency was strongly associated with increased disease severity and adverse clinical outcomes in pediatric inpatients with ARI.

KEYWORDS: Vitamin D deficiency, acute respiratory infections, disease severity, pediatrics.

Introduction

Acute respiratory infections (ARIs) encompass a broad spectrum of conditions affecting the upper and lower respiratory tract.

Upper respiratory tract infections involve the airways from the nostrils to the vocal cords, including the paranasal sinuses and middle ear, whereas lower respiratory tract infections extend from the trachea and bronchi to the bronchioles and alveoli [1].

ARIs represent one of the leading causes of morbidity and hospitalization in children worldwide, with clinical presentations ranging from mild self-limited illness to severe lower respiratory tract disease, such as bronchiolitis and pneumonia [1,2].

Although ARIs primarily affect the respiratory system, they may also produce systemic effects mediated by inflammation, microbial toxins, and impaired pulmonary function, thereby contributing to increased disease severity and adverse clinical outcomes [1].

These systemic manifestations play an important role in determining the clinical course and prognosis of pediatric ARI.

In recent previous years, increasing attention had been directed toward the role of vitamin D in immune regulation and respiratory health.

Vitamin D exerts key immunomodulatory effects by enhancing innate immune responses, promoting antimicrobial peptide synthesis, and regulating adaptive immunity and inflammatory pathways [3].

A growing body of pediatric evidence indicates that low serum 25-hydroxyvitamin D [25(OH)D] levels are associated with an increased risk and severity of acute respiratory infections, including viral respiratory infections, bronchiolitis, and community-acquired pneumonia [4-9].

Several studies have demonstrated that infants and young children with vitamin D deficiency experience more severe infection of lower respiratory tract, accompanied by heightened inflammatory responses, prolonged length of hospitalization, and increased need for respiratory support [4-9].

Despite these consistent findings, data from Eastern European pediatric populations, particularly from Romania, remain limited [10].

A clearer understanding of the relationship between vitamin D status and ARI severity is

essential for improving risk stratification and clinical management in hospitalized children.

Therefore, the present study aimed to assess the prevalence of vitamin D deficiency among pediatric patients hospitalized with acute respiratory infections and to investigate its association with disease severity, inflammatory biomarkers, and key clinical outcomes, including fever, oxygen therapy, and length of hospital stay.

Material and Methods

Study Design and Setting

This observational case-control study 1:1 was conducted over a four-year period (2021-2024) in two pediatric healthcare institutions, placed in South Eastern Romania: Medgidia Municipal Hospital and the Pediatric Clinic of the Constanța Clinical Emergency County Hospital, a tertiary referral center serving both urban and rural populations.

During the study period, all consecutive pediatric patients aged 0-18 years, who were hospitalized with a clinically confirmed acute respiratory infection (ARI), were prospectively evaluated.

The spectrum of ARIs included upper respiratory tract infections (acute laryngitis, acute pharyngitis, and acute otitis media), or lower respiratory tract infections (bronchitis, bronchiolitis, community-acquired pneumonia, bronchopneumonia), inclusive laboratory-confirmed COVID-19 infection.

All diagnostic categories were systematically recorded for each enrolled patient.

Upon admission, all eligible patients underwent standardized clinical assessment and routine laboratory investigations, including measurement of serum 25-hydroxyvitamin D [25(OH)D] levels by chemiluminescence immunoassay, respecting previous daily internal quality control procedures.

The study was approved by the Ethics Committee of Medgidia Municipal Hospital (approval no. 152/06.01.2022) and by the Ethics Committee of the "Sf. Apostol Andrei" County Emergency Hospital, Constanța (approval no. 2770/16.01.2024).

Written informed consent was mandatory to be obtained from all parents, or legal guardians prior to enrollment.

A total of 400 pediatric patients aged 0-18 years who met the predefined inclusion criteria: (1) clinically confirmed ARI and (2) availability of serum vitamin D assessment.

Exclusion criteria included chronic kidney or liver disease, cystic fibrosis, autoimmune

disorders, malignancies, and immunodeficiency syndromes that could independently affect vitamin D metabolism or interfere the hospitalization duration.

Patients were stratified according to vitamin D status into two equal groups:

- Cases with low level of vitamin D (n=200): serum 25(OH)D levels <30ng/mL, including vitamin D deficiency (<20ng/mL) and insufficiency (20-29ng/mL), based on WHO-aligned criteria [11].

For analytical purposes, deficiency and insufficiency were analyzed together.

- Controls with normal vitamin D (n=200): serum 25(OH)D levels ≥30ng/mL.

The diagnosis of ARI was established based on clinical evaluation, supported by inflammatory laboratory markers and virological testing, when indicated [12].

Age categories were defined according to World Health Organization pediatric age groups: infants (0-1 year), young children (1-4 years), older children (5-9 years), and adolescents (10-18 years) [13].

For statistical analysis in SPSS, all children younger than 1 year were coded as 1 year of age; therefore, the age category of 1 year in the graphical representations includes all patients aged under 1 year.

Clinical and Laboratory Data Collection

For each eligible subject, clinical and laboratory data were collected at admission using a standardized data collection form.

Recorded variables included demographic characteristics (age, sex, place of residence, and season of hospitalization), clinical parameters (maximum documented body temperature, requirement for supplemental oxygen, duration of hospital stay, and clinician-rated disease severity), and laboratory measurements of serum 25(OH)D concentration, C-reactive protein (CRP) as a marker of systemic inflammation, total immunoglobulin E (IgE), and hemoglobin levels.

All laboratory analyses, performed in two different hospitals, used the same certified analytical method.

Outcome Measurements

The primary outcome was ARI disease severity, categorized as mild, moderate, or severe.

To provide a standardized assessment of illness severity, a **composite clinical severity score**, ranging from 0 to 7 points, was developed

by the authors, incorporating four clinical and paraclinical parameters:

- **Length of hospital stay:**
1-3 days=0 points
4-6 days=1 point
≥7 days=2 points
- **C-reactive protein (CRP):**
≤1 mg/dL=0 points
1.1-4.9 mg/dL=1 point
≥ 5 mg/dL=2 points
- **Fever:**
Present=1 point
Absent=0 points
- **Oxygen therapy:**
Required=2 points
Not required=0 points

The total score was calculated by summing all components (possible range: 0-7 points).

Based on the composite clinical severity score, patients were classified as having different stage of **Disease Severity Classification:**

- **Mild disease:** 0-2 points,
- **Moderate disease:** 3-5 points,
- **Severe disease:** 6-7 points.

This classification provided an integrated evaluation of respiratory compromise, systemic inflammation, and overall clinical status.

Statistical Analysis

Continuous variables were expressed as mean±standard deviation or median with

interquartile range, depending on data distribution assessed by normality testing.

Comparisons between groups were performed using independent-samples t-tests for normally distributed variables, while appropriate non-parametric tests were applied for non-normally distributed data.

Categorical variables were presented as frequencies and percentages and compared using chi-square or Fisher’s exact tests, as appropriate.

Associations between vitamin D status and clinical outcomes-including severe ARI, presence of fever, and requirement for oxygen therapy-were evaluated using odds ratios (ORs) with 95% confidence intervals (CIs).

Multivariate logistic regression models were considered to adjust for potential confounders, including age, season of hospitalization, and place of residence.

A p-value <0.05 was considered statistically significant. All analyses were performed using standard statistical software.

Results

Baseline Characteristics

A total of 400 pediatric patients aged 0-18 years were included in the analysis, comprising 200 cases with low serum vitamin D levels, and 200 controls with normal vitamin D levels.

Baseline demographic and clinical characteristics are summarized in Table 1.

Table 1. Baseline characteristics of pediatric patients according to vitamin D status.

| Variable | Cases (n=200) | Controls (n=200) |
|--------------------------------|---------------|------------------|
| Age (years) | 5.79±4.73 | 4.43±4.37 |
| Sex (% female) | 41.0% | 48.5% |
| Residence (% urban) | 32.5% | 33.5% |
| Hospital length of stay (days) | 4.68±2.59 | 2.89±1.81 |
| Vitamin D (ng/mL) | 21.62±5.56 | 47.60±19.59 |
| CRP (mg/L) | 3.50±3.02 | 1.64±1.59 |
| Clinical severity score | 3.77±2.29 | 1.62±1.89 |
| Hemoglobin (g/dL) | 11.45±1.78 | 12.42±1.26 |

Legend: CRP=C-reactive protein

The mean age was slightly higher in the low vitamin D group compared with controls (5.79±4.73 vs. 4.43±4.37 years).

Serum 25-hydroxyvitamin D concentrations confirmed appropriate group allocation, with significantly lower levels in the low vitamin D group than in controls (21.62±5.56ng/mL vs. 47.60±19.59ng/mL).

In cases group, one-way ANOVA showed no statistically significant differences in gender distribution ($F=0.873$, $p<0.595$) or residence

($F=1.628$, $p<0.070$) across different age (Table 2).

Similarly, in controls group, no significant differences were identified by gender ($F=1.446$, $p<0.136$) or residence ($F=0.992$, $p<0.464$) across age categories (Table 2).

Additional analyses assessing linear trends and deviations from linearity were non-significant in both cases and controls (all $p>0.05$).

Table 2. One-way ANOVA results for gender and place of residence among cases and controls by vitamin D status.

| Vitamin D group | Variable | F | df | p< |
|-----------------|-----------|-------|--------|-------|
| Low | Gender | 0.873 | 15,184 | 0.595 |
| Low | Residence | 1.628 | 15,184 | 0.070 |
| Sufficient | Gender | 1.446 | 14,185 | 0.136 |
| Sufficient | Residence | 0.992 | 14,185 | 0.464 |

Legend: df= degree of freedom

Figures 1 and 2 illustrate the distribution of male and female patients across age groups in the low and normal vitamin D groups, respectively.

In both groups, boys and girls were represented across all age categories, with some

variation in absolute numbers between age groups.

Higher counts were observed in the youngest age category, reflecting the age structure of the study population.

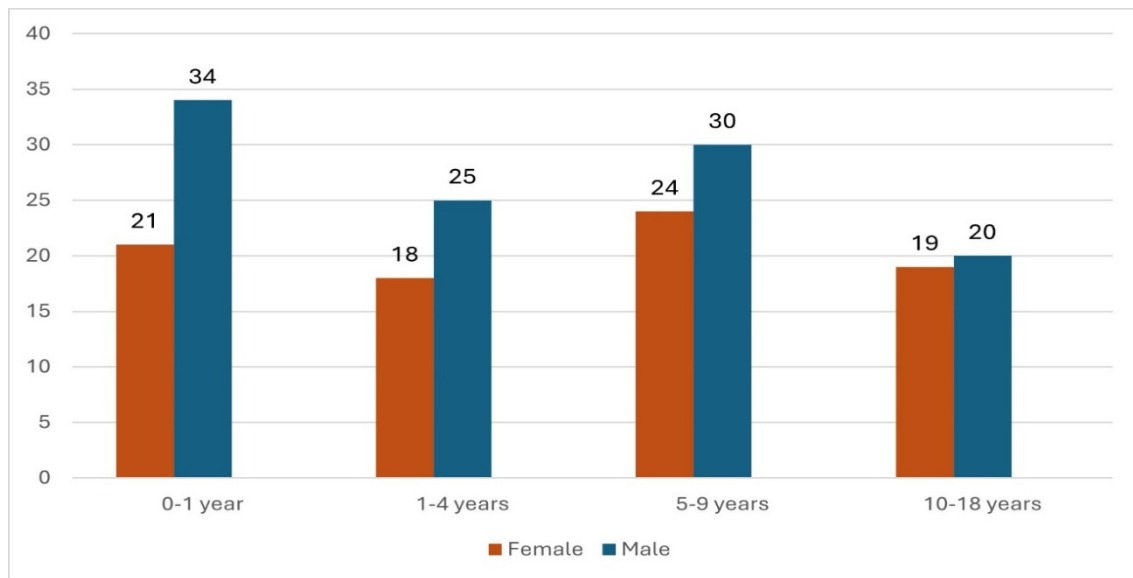


Figure 1. Distribution of cases (n=200) by gender and age.

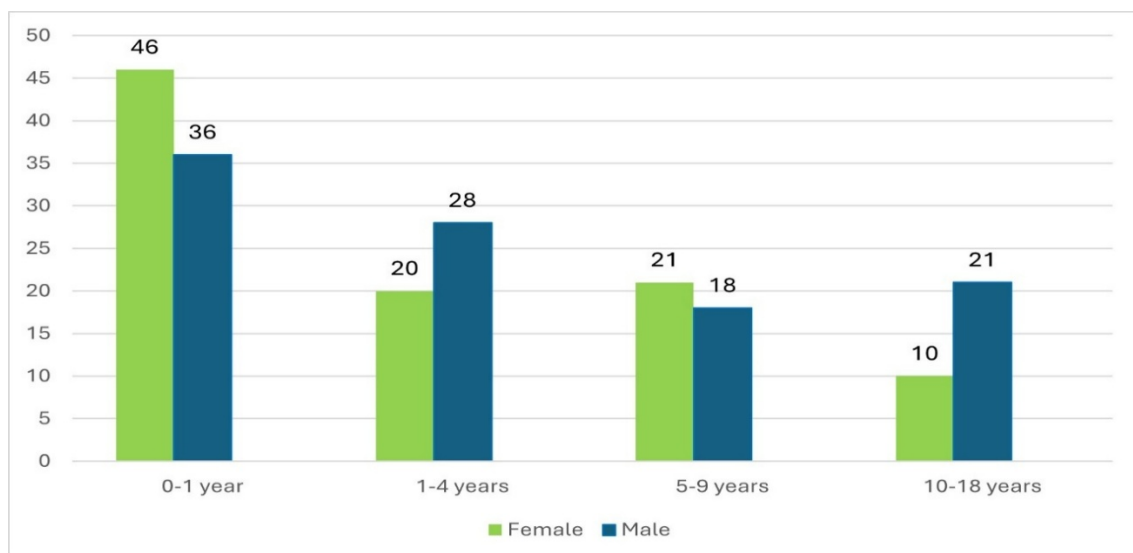


Figure 2. Distribution of controls (n=200) by gender and age.

However, no consistent age-related pattern in sex distribution was evident, in agreement with the results of the ANOVA analyses, which

showed no statistically significant by gender and age distribution.

Inflammatory Biomarkers and Clinical Severity Indicators of Acute Respiratory Infections

Significant differences were observed between groups with respect to inflammatory and severity-related parameters (Table 1).

Cases had statistically significant higher C-reactive protein concentrations compared with controls (3.50±3.02mg/L vs. 1.64±1.59mg/L; $p<0.001$).

The composite clinical severity score was significantly higher in the low vitamin D group (3.77±2.29) than in patients with normal vitamin D levels (1.62±1.89; $p<0.001$).

Hemoglobin levels were significantly lower in cases than in controls (11.45±1.78/dL vs. 12.42±1.26g/dL; $p<0.001$).

No statistically significant differences were observed in total IgE concentrations between groups.

Disease Severity and Clinical Outcomes of Acute Respiratory Infections

The length of hospital stay was significantly longer in patients with low vitamin D levels compared with controls (4.68±2.59 vs. 2.89±1.81 days; $p<0.001$) (Table 1).

A marked difference in disease severity distribution was observed between groups (Table 3).

Table 3. Disease severity and clinical outcomes of acute respiratory infections among cases and controls according to vitamin D status.

| Variable | Category | Cases (n=200) | Controls (n=200) |
|------------------|----------|---------------|------------------|
| Disease severity | Mild | 63 (31.5%) | 145 (72.5%) |
| | Moderate | 84 (42.0%) | 44 (22.0%) |
| | Severe | 53 (26.5%) | 11 (5.5%) |
| Fever | Yes | 122 (61.0%) | 64 (32.0%) |
| | No | 61 (39.0%) | 136 (68.0%) |
| Oxygen therapy | Yes | 139 (69.5%) | 42 (21.0%) |
| | No | 61 (30.5%) | 158 (79.0%) |

Severe acute respiratory infections (ARIs) occurred significantly more frequently in cases (26.5%) than in the control group (5.5%) (OR=6.20, 95% CI: 3.13-12.27; $p<0.001$), while mild disease predominated among controls (72.5% vs. 31.5%; $p<0.001$).

Fever at admission was more common among patients with low vitamin D levels, affecting 61.0% of cases compared with 32.0% of controls (OR=11.59, 95% CI: 7.14-18.81; $p<0.001$).

Similarly, the need for supplemental oxygen therapy was significantly higher in the low vitamin D group (69.5%) than in the control group (21.0%) (OR=5.88, 95% CI: 3.78-9.17; $p<0.001$).

Discussion

In this prospective 4-year case-control study involving 400 pediatric patients hospitalized with acute respiratory infection (ARI), low serum levels of 25-hydroxyvitamin D were strongly associated with increased disease severity and adverse clinical outcomes.

Patients with vitamin D deficiency (VDD) exhibited higher rates of severe ARI, more frequent fever, increased need for oxygen therapy, elevated inflammatory markers, and prolonged length of hospitalization compared with those with normal vitamin D levels.

These findings are consistent with a growing body of pediatric literature demonstrating an association between VDD and more severe ARIs, including bronchiolitis, pneumonia, and other lower respiratory tract infections [4-7,14-18].

Data from hospitalized pediatric patients with viral etiology of ARI (including pneumonia and COVID-19 infection) further support this relationship, linking low vitamin D levels to increased inflammatory burden, longer hospital stays, and greater respiratory support requirements [8,9,19].

The consistency of these association across different respiratory pathogens suggests a broader immunomodulatory role of vitamin D in respiratory infections.

The biological plausibility of severe ARI-VDD related is supported by mechanistic evidence indicating that vitamin D enhances epithelial barrier function, induces antimicrobial peptides such as cathelicidin (LL-37) and β -defensins, and modulates immune responses by attenuating excessive pro-inflammatory cytokine production while promoting immune regulation [20].

These effects may contribute to the more pronounced systemic inflammation observed in vitamin D-deficient patients in the present study, as reflected by higher C-reactive protein (CRP)

levels and elevated clinical severity scores of ARI.

Similar inflammatory profiles have been reported in other pediatric studies linking vitamin D deficiency with heightened cytokine responses and more severe pulmonary involvement [14,17,18,21-23].

Our results further reinforce evidence from prospective and case-control studies identifying VDD as an independent predictor of poor outcomes in pediatric lower respiratory tract infections, including prolonged hospitalization, hypoxia, and severe disease [14,17,18,22].

Notably, a recent systematic review and meta-analysis published in 2024 concluded that VDD represents one of the most consistent risk factors for severe ARI across pediatric populations worldwide [23].

Together, these findings underscore the relevance of vitamin D status in clinical risk stratification.

The strengths of this study include the relatively large sample size, prospective data collection, use of a well-defined control group, and assessment of multiple clinically relevant outcomes.

Measurement of serum vitamin D levels at admission minimized the potential influence of in-hospital interventions on vitamin D status.

In our study, VDD was associated with more than a six-fold increase in the odds of severe ARI and oxygen requirement, and with substantially prolonged hospitalization, representing strong and clinically meaningful associations.

Several limitations should be acknowledged.

The observational design precludes causal inference, as severe illness may secondarily influence vitamin D metabolism.

The study was conducted in two hospitals within a single geographical region, potentially limiting generalizability.

Additionally, the heterogeneous spectrum of respiratory infections and the absence of data on nutritional status, sun exposure, socioeconomic factors, or prior vitamin D supplementation may have introduced residual confounding.

Despite these limitations, the consistency and magnitude of the observed associations support the conclusion that low vitamin D status is strongly linked to increased severity of acute respiratory infections in children.

From a clinical perspective, early identification of vitamin D deficiency may help identify pediatric patients at higher risk for severe disease and guide closer monitoring.

Further multicenter, prospective, and interventional studies are warranted to determine whether optimization of vitamin D status can improve outcomes and reduce the burden of pediatric ARIs.

Conclusions

Vitamin D deficiency was strongly associated with increased disease severity of acute respiratory infections (ARIs) and poorer clinical outcomes in hospitalized pediatric patients.

Low serum 25-hydroxyvitamin D levels were linked to greater systemic inflammation, higher frequency of fever, increased need for oxygen therapy, and prolonged hospitalization.

These findings support the potential role of vitamin D status as a prognostic biomarker of severe pediatric ARI and underscore its relevance for clinical risk assessment.

Further prospective and interventional studies are needed to determine whether optimizing vitamin D levels can improve outcomes in pediatric respiratory infections.

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None to declare.

Author Contributions

M.N. contributed to data collection, data analysis, and manuscript drafting. T.C. and C.M.M. participated in data acquisition and interpretation. O.C.A. contributed to the conception and design of the study. M.N. was involved in data analysis. C.M.M. and O.C.A. contributed to statistical analysis and interpretation of the results. T.C. participated in manuscript review and editing. C.M.M. and O.C.A. supervised the study and provided critical revision of the manuscript.

All authors read and approved the final manuscript.

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Conflicts of Interest

The authors declare no competing interests.

Institutional Review Board

All procedures followed were in accordance with the ethical standards of the responsible committee for human studies (institutional and national) and with the latest revision of the Helsinki declaration.

The study was approved by the Ethics Committee of Medgidia Municipal Hospital (approval no. 152/06.01.2022) and by the Ethics Committee of the "Sf. Apostol Andrei" County Emergency Hospital, Constanța (approval no. 2770/16.01.2024).

Consent Statement

All human subjects involved in this study signed institutional consent forms upon hospital admission allowing the use of their anonymized medical data for research purposes.

Data Availability

All data presented in this manuscript are available from the corresponding author upon reasonable request.

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