Vitamin D in Pediatric Inpatients With Respiratory Illnesses

abstract

BACKGROUND AND OBJECTIVE: Low serum 25-hydroxyvitamin D (25(OH)D) levels have been associated with increased susceptibility to and severity of respiratory viral infections. Hypovitaminosis D may be a modifiable risk factor in the severity of viral respiratory illnesses. The hypothesis for this study was that children hospitalized for respiratory illnesses would have lower serum 25(OH)D levels than controls and that 25(OH)D levels would be associated with illness severity among cases.

METHODS: A case–control study of a sample of patients aged 6 months through 12 years hospitalized from January to May 2010 at an urban pediatric referral hospital was performed. Cases were children hospitalized for acute respiratory illnesses, and controls were children hospitalized for nonrespiratory illnesses. Illness severity among cases was assessed according to hospital length of stay, ICU admission, peripheral oxygen saturation, and pediatric risk of admission II score. Associations between serum 25(OH)D levels and dependent variables were tested for by using binary logistic and multivariable linear regression while controlling for admission diagnosis, age, gender, and race/ethnicity.

RESULTS: The majority of cases (n = 38) and controls (n = 83) were African American (65.8% and 59.0%, respectively). Of the entire cohort (N = 121), 64.8% had vitamin D insufficiency (25(OH)D level ≤30 ng/mL) and 31.1% had vitamin D deficiency (25(OH)D level ≤20 ng/mL). Mean ± SD 25(OH)D levels did not differ between cases and controls (26.8 ± 11.5 vs 26.1 ± 10.6 ng/mL, respectively; P = .73).

CONCLUSIONS: Hypovitaminosis D was common among cases and controls, but it was not significantly associated with the presence or severity of respiratory illnesses.

Vitamin D is a steroid hormone synthesized in the skin through sunlight exposure or ingested through the diet. It is typically measured as serum 25-hydroxyvitamin D (25(OH)D). In its active form, 1,25-dihydroxyvitamin D (1,25(OH)D) is involved in defense against viral, bacterial, and mycobacterial infections in pulmonary tissues and in down-regulating inflammation by decreasing chemokine and cytokine production. The antiinflammatory actions of 1,25(OH)D have also been shown in in vitro models of lung tissue; when inflamed airway smooth muscle cells are treated with increasing doses of 1,25(OH)D, they exhibit dose-dependent decreases in the production of inflammatory cytokines.

Vitamin D insufficiency (25(OH)D serum level ≤30 ng/mL) and deficiency (25(OH)D level ≤20 ng/mL) are widespread throughout multiple populations and
are especially common in African-American individuals, partly due to the amount of melanin in the skin. Melanin blocks UV radiation and decreases production of vitamin D. Other factors in vitamin D deficiency or insufficiency include low vitamin D intake, urban residence, poverty, and obesity. Vitamin D levels vary throughout the year; studies have shown that darker-skinned children living in the Northeast have uniformly low 25(OH)D levels in the winter months.

Low 25(OH)D levels have been associated with increased severity of immune-mediated diseases such as asthma. This finding is thought to be at least partly due to increased susceptibility to viral infections. Associations between asthma development/severity and low prenatal and early childhood serum vitamin D levels have also been well established in pediatric populations. In addition, 25(OH)D deficiency and insufficiency have been shown to be associated with increased susceptibility to and increased severity of respiratory illnesses in children.

In Japan, Inamo et al found that pediatric inpatients with respiratory illnesses had more severe courses if they were 25(OH)D deficient, and in Bangladesh, Roth et al showed that children admitted for acute lower respiratory tract infections to a general inpatient ward had lower 25(OH)D levels than age-matched healthy controls. Furthering the association between severity of respiratory illnesses and vitamin D sufficiency, a randomized, double-blind, controlled trial of vitamin D supplementation in children in Kabul showed that supplementation with vitamin D after a diagnosis of pneumonia was associated with fewer pneumonia recurrences within the first 90 days after primary illness.

Contrary to those findings, McNally et al reported no differences in serum 25(OH)D levels between 105 cases of pediatric inpatients with bronchiolitis and pneumonia and those of 92 controls, but they did find an association between lower 25(OH)D levels and increased ICU admission. In Canada, Roth et al also reported that no associations existed between vitamin D levels and need for admission in children <2 years of age diagnosed with bronchiolitis.

Given the evolving understanding of 1,25(OH)D in the role of respiratory health and the potential of low vitamin D levels to be associated with increasing severity of acute respiratory illnesses, the goal of the current study was to compare serum 25(OH)D levels among hospitalized children with and without respiratory illnesses. The primary hypothesis was that low serum 25(OH)D levels would be more common among children hospitalized with respiratory illnesses than among children hospitalized for other reasons. A further hypothesis was that low serum 25(OH)D levels would be associated with increased severity of disease among those patients hospitalized with respiratory illnesses.

**METHODS**

**Study Design**

This study was a case–control trial of patients aged 6 months through 12 years hospitalized at Children’s National Medical Center in Washington, DC (latitude 38°/longitude 55°), from January 1, 2010, through May 1, 2010. All children were from the Washington, DC, metropolitan area. All children admitted from the emergency department aged 6 months to 12 years who had blood drawn on admission were eligible for inclusion in the study. Exclusion criteria included being in foster care; having a known significant illness that affects vitamin D levels; having a chronic pulmonary disease other than asthma (including but not limited to cystic fibrosis and bronchopulmonary dysplasia); having renal, cardiac, or gastrointestinal disease; receiving vitamin D supplementation; or not speaking English or Spanish.

Cases (n = 38) included children hospitalized with a primary admission diagnosis of an acute respiratory illnesses (including, but not limited to, asthma, pneumonia, and bronchiolitis). Controls (n = 83) included children admitted with a primary admission diagnosis of a nonrespiratory illnesses (diagnoses not attributable to a primary pulmonary issue). All primary admission diagnoses were compared with discharge diagnoses to ensure that no vitamin D–related illness was diagnosed during the course of the child’s stay, and 1 child was removed from the control group. He had a primary admission diagnosis of an apparent life-threatening event later found to be a hypocalcemic seizure due to vitamin D deficiency. Each child was enrolled only once during the study period, and none of the control patients was subsequently hospitalized for a respiratory illness during the study period at the study institution. 25(OH)D levels were compared between cases and controls.

Cases were also secondarily analyzed in a cross-sectional manner to determine if 25(OH)D levels were related to the following clinical outcomes: hospital length of stay (LOS), admission to the PICU, magnitude of hypoxia (defined as lowest recorded peripheral oxygen saturation), and pediatric risk of admission II (PRISA II) score. PRISA II is a
validated pediatric measure that was designed to determine risk of inpatient admission on presentation to the pediatric emergency department. This value allowed categorization of patients on the basis of initial illness severity. The variables included in PRISA II are: age, baseline immunodeficiency, indwelling medical device, controller asthma medications, temperature, mental status, blood pressure, need for oxygen, serum bicarbonate, potassium, blood urea nitrogen, and white blood cell count.

Guardians of patients who agreed to participate in the study were asked to identify their child’s race, ethnicity, and presence or absence of clinician-diagnosed asthma for >1 year. The institutional review board at Children’s National Medical Center approved this study.

**Vitamin D Measurements**

25(OH)D levels were determined by using surplus plasma or serum available in the clinical laboratory. Serum aliquots for each enrolled subject were shipped on dry ice to Massachusetts General Hospital (Boston, MA) for serum 25(OH)D measurement according to liquid chromatography–tandem mass spectrometry (LC-MS). This method uses an isotope dilution, LC-MS assay optimized in the hospital laboratory. The lower limits of detection by LC-MS are 5 nmol/L for vitamin D$_2$ (ergocalciferol) and 25 nmol/L for vitamin D$_3$ (cholecalciferol). The between-run coefficient of variation for a quality control serum containing a total vitamin D concentration of 57 nmol/L is 7.5%. Total vitamin D$_2$ and D$_3$ measurements were combined to give final vitamin D concentrations and converted nanomoles per liter into nanograms per milliliter. Vitamin D insufficiency was defined as a 25(OH)D level ≤30 ng/mL, and vitamin D deficiency was defined as a 25(OH)D level ≤20 ng/mL.2728

**Data Abstraction**

All charts of cases were reviewed for markers of disease severity (LOS, admission to PICU, and hypoxia). Variables for the PRISA II score26 were determined by analysis of the emergency department electronic medical record by a research assistant who entered information on a score sheet. On completion of the score sheet, the research assistant calculated the PRISA II score. The primary investigator oversaw both score sheets and composite scores for all PRISA II scoring, and in the case of disputed scores, her score was the one used.

**Data Analysis**

Descriptive statistics and frequencies were analyzed by using Student’s $t$ tests and $\chi^2$ tests. Two-tailed $P$ values ≤.05 were considered significant, and all reported confidence intervals (CIs) were at 95%. Associations between serum 25(OH)D and chosen dependent variables were tested by using binary logistic regression (for PICU admission) and multivariable linear regression (for PRISA II scores, LOS, and hypoxia). Odds ratios, $\beta$ coefficient ($\beta$), and $P$ values were corrected for age, gender, race, and ethnicity. Variability in the chosen outcome variables of LOS, hypoxia, and PRISA II attributable to 25(OH)D were determined by performing linear regression on two different models (with and without 25(OH)D). Difference in variability attributable to 25(OH)D was determined by the difference in the $R^2$ between the 2 models. All statistical tests were performed by using SPSS version 17.0 (IBM SPSS Statistics, IBM Corporation, Armonk, NY).

**Sample Size**

In a previous study published in 2011, 54% of African American patients with asthma aged 6 to 20 years from the metropolitan Washington, DC, area were vitamin D deficient.29 Using this proportion as a proxy for vitamin D deficiency among the cases, a sample size of 41 in each group gave a power of .80 in a 2-sided test with an $\alpha$ level of .05 to detect a 30% difference in the proportion of patients with 25(OH)D deficiency between groups.

**RESULTS**

**Population**

Of the 677 eligible patients admitted during the study period, 167 had excess blood in the laboratory for vitamin D testing. Of these, 3 families refused to participate, and 22 families could not be contacted for consent. An additional 20 children (4 cases and 16 controls) had insufficient blood volume to run a 25(OH)D level, and 1 additional child had to be removed from controls for a discharge diagnosis of vitamin D deficiency leaving 38 cases and 83 controls for analysis (Fig 1). Cases were significantly older than controls and were significantly more likely to have a previous diagnosis of asthma (47.4% vs 14.5%; $P = .01$). Cases and controls were otherwise similar with respect to gender, race, and ethnicity in unadjusted univariate analysis (Table 1). Children admitted to the PICU tended to be younger and more likely to have asthma than children who did not require PICU admission (Table 2). Mean vitamin D levels were significantly lower in PICU patients than in patients admitted to the general pediatric services; however, when controlling for age, gender, race, and ethnicity, this difference disappeared.

The 38 cases consisted of patients hospitalized with pneumonia ($n = 10$), asthma exacerbations without identified
concomitant pneumonia \((n = 9)\), asthma exacerbations with concomitant pneumonia \((n = 7)\), bronchiolitis \((n = 4)\), other illnesses \((n = 4)\), respiratory distress/failure \((n = 3)\), and empyema \((n = 1)\). Serum 25(OH)D levels did not vary by admission diagnosis. Respiratory viral polymerase chain reaction was performed in all but 9 of the children. Of the positive results, 8 patients had human metapneumovirus, 8 had rhinovirus/enterovirus, 5 had respiratory syncytial virus, 3 had adenovirus, and 1 had parainfluenza. One test yielded an inconclusive result, and 3 had negative results. Among cases, the median PRISA II score was 28 (range: 3–84).

The 83 controls were patients admitted to the hospital with nonrespiratory diagnoses. Admission diagnoses were as follows: skin infections \((n = 11)\), hyperglycemia \((n = 10)\), appendicitis \((n = 8)\), seizure \((n = 7)\), closed head injury \((n = 6)\), Kawasaki disease \((n = 6)\), fracture \((n = 5)\), and fever \((n = 3)\). Two patients each had gastroenteritis, accidental ingestions, abdominal trauma, lymphadenitis, deep neck abscesses, and bone infections. The remaining 15 controls had admission diagnoses including dysfunctional uterine bleeding, ovarian cyst, ovarian torsion, mid-gut volvulus, acute ataxia, smoke inhalation, anemia, pulmonary contusion, abdominal pain of unknown etiology, hematuria, encephalitis, pectus excavatum, pancreatitis, thrombocytopenia, and febrile seizure.

**Vitamin D Levels**

Vitamin D levels were normally distributed among our population: the mean vitamin D level was 26.6 ng/mL and the median vitamin D level was 26.3 ng/mL. Skewness among the group as a whole was 0.404 (SE: 0.2), and kurtosis was −0.22 (SE: 0.47); thus, our data were neither significantly skewed nor kurtotic. Skewness was 0.13 (SE: 0.38) among cases and 0.56 (SE: 0.26) among controls. Kurtosis was −0.85 (SE: 0.75) among cases and 0.23 (SE: 0.52) among controls. Using an independent sample \(t\) test, the means for cases and controls were, respectively, 26.8 \(±\) 11.5 vs 26.1 \(±\) 10.6 ng/mL \((P = .73)\) [95% confidence interval: −3.5 to 4.9].

**Case-Control Study**

For the entire cohort, 64.8% of all children were vitamin D insufficient and 31.1% were vitamin D deficient. The proportions of cases and controls who were vitamin D insufficient (57.9% vs 68.7%, respectively; \(P = .30\)) and vitamin D deficient (34.2% vs 30%, respectively; \(P = .67\)) did not differ significantly between groups. Mean serum 25(OH)D levels also did not differ between cases and controls (26.8 \(±\) 11.5 vs 26.1 \(±\) 10.6 ng/mL, respectively; \(P = .73\)). Across both cases and controls, an increase

**TABLE 1 Comparison of Cases and Controls**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Cases ((n = 38))</th>
<th>Controls ((n = 83))</th>
<th>(P^a)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, mean (±) SD, y</td>
<td>8.6 (±) 7.5</td>
<td>5.4 (±) 3.8</td>
<td>.002</td>
</tr>
<tr>
<td>Male gender</td>
<td>20 (52.6)</td>
<td>43 (51.8)</td>
<td>.933</td>
</tr>
<tr>
<td>Race</td>
<td></td>
<td></td>
<td>.544</td>
</tr>
<tr>
<td>African American</td>
<td>25 (65.8)</td>
<td>49 (59.0)</td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>3 (7.9)</td>
<td>15 (18.1)</td>
<td></td>
</tr>
<tr>
<td>Asian</td>
<td>1 (2.6)</td>
<td>2 (2.4)</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>9 (23.7)</td>
<td>17 (20.5)</td>
<td></td>
</tr>
<tr>
<td>Ethnicity</td>
<td></td>
<td></td>
<td>.937</td>
</tr>
<tr>
<td>Hispanic</td>
<td>8 (21.1)</td>
<td>18 (21.7)</td>
<td></td>
</tr>
<tr>
<td>Non-Hispanic</td>
<td>30 (78.9)</td>
<td>65 (78.3)</td>
<td></td>
</tr>
</tbody>
</table>

Data are presented as mean \(±\) SD or \(n\) (%).

* All \(P\) values reflect unadjusted univariate analysis by Student’s \(t\) tests or \(\chi^2\) tests.
of 1 year in age was associated with a 0.92 ± 0.16 decrease in 25(OH)D (P < .001). These results remained consistent even when all patients with asthma were removed from the control group. When we removed all patients with primary admission diagnoses of infectious processes from the control group, mean serum 25(OH)D levels did not differ between cases and controls (26.8 ± 11.5 vs 26 ± 11.2 ng/mL, respectively; P = .72), and the proportions of cases and controls who were vitamin D insufficient (57.0% of cases and 68.6% of controls; P = .37) and deficient (34.2% of cases and 29.0% of controls; P = .65) also did not differ significantly.

Cross-Sectional Study
Among cases, serum 25(OH)D levels were not significantly associated with hospital LOS, hypoxia, or PRISA II score when adjusting for admission diagnosis, age, gender, race, and ethnicity. In fact, serum 25(OH)D was minimally responsible for variation within our 3 outcomes (Table 3). Decreased serum vitamin D levels were not associated with increased odds of admission to the PICU when adjusted for admission diagnosis, age, gender, race, or ethnicity (odds ratio: 0.93 [95% confidence interval: 0.84 to 1.03], P = .14).

Discussion
In this case–control study of hospitalized patients aged 6 months to 12 years with and without respiratory illnesses, the majority of both the case and control groups had vitamin D insufficiency. Neither the proportion of patients with vitamin D deficiency nor mean serum levels differed by group. Among cases, there were no associations between vitamin D level and illness severity, as defined by hospital LOS, need for PICU admission, PRISA II score, or degree of hypoxia.

This study had several limitations. First, the final number of cases eligible for inclusion failed to meet the projected sample size. Nonetheless, when power was recalculated based on the actual sample size, there was 80% power to detect a difference of 6.3 ng/mL in 25(OH)D levels and 90% power to detect a difference of 7.3 ng/mL. Previous studies have shown that 25(OH)D deficiency is not exceptionally high in the urban Washington, DC, population and have also shown a wide variability within 25(OH)D levels: median yearly 25(OH)D in healthy African-American children living in Washington, DC, was 40.1 ng/mL and median 25(OH)D in African-American children with asthma was 18.5 ng/mL. Mean vitamin D levels in the cases and controls in the current study fell roughly between these 2 numbers. The results showed no clear trend toward a difference in 25(OH)D levels and, therefore, a modest increase in the number of cases would be unlikely to affect these findings.

Second, the case subjects had a variety of different respiratory disorders, and it is possible that low 25(OH)D levels might be associated with one of the disorders when individually compared with controls. Future studies to examine vitamin D among respiratory illnesses would benefit from focusing on a single illness. Third, we were not able to control for travel before hospitalization; however, our population is largely urban and underserved, and travel to areas of increased sun exposure is likely to be limited. Fourth, it is possible that 25(OH)D levels might be associated with acute respiratory illnesses in children with sufficient 25(OH)D levels; conducting this study in the summer, when vitamin D levels are naturally at their highest, might reveal such a difference. Finally, these results were drawn from a population of largely urban and minority children, and they may not be generalizable to other populations.

Although many studies have shown associations between vitamin D deficiency and respiratory illnesses, our study found no such difference. Negative studies add to the general body of knowledge regarding vitamin D, and they indicate that vitamin D alone may not have dramatic

Table 2: Comparison of PICU Admissions With General Hospital Admissions

<table>
<thead>
<tr>
<th>Variable</th>
<th>PICU Admissions (n = 12)</th>
<th>Hospital Admissions (n = 26)</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean vitamin D levels</td>
<td>19.2</td>
<td>30.4</td>
<td>.001</td>
</tr>
<tr>
<td>Age, y</td>
<td>6.3</td>
<td>13.5</td>
<td>.03</td>
</tr>
<tr>
<td>% with asthma diagnosis</td>
<td>66.7% (n = 8)</td>
<td>38.5% (n = 10)</td>
<td>.11</td>
</tr>
</tbody>
</table>

*All P values reflect unadjusted univariate analysis by Student’s t tests or χ² tests.

Table 3: Associations of Vitamin D With Various Outcome Variables While Controlling for Admission Diagnosis, Age, Gender, Race, and Ethnicity

<table>
<thead>
<tr>
<th>Variable</th>
<th>Coefficient for 25(OH)D</th>
<th>95% CI</th>
<th>P</th>
<th>Variability Attributable to 25(OH)D (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hospital LOS</td>
<td>−0.08</td>
<td>−0.23 to 0.7</td>
<td>.29</td>
<td>0.02</td>
</tr>
<tr>
<td>Degree of hypoxia</td>
<td>−144.3</td>
<td>−6278 to 339</td>
<td>.54</td>
<td>1.1</td>
</tr>
<tr>
<td>PRISA II</td>
<td>−0.23</td>
<td>−0.84 to 0.37</td>
<td>.44</td>
<td>1.4</td>
</tr>
</tbody>
</table>

The difference in variability attributable was determined by the difference in the R² between linear regression models including and not including 25(OH)D as a predictive factor. CI, confidence interval.
effects on acute respiratory illnesses in cohorts similar to ours. Instead, it is more likely that vitamin D is 1 of multiple nutritional exposures that affect respiratory health. Future studies of vitamin D and acute illness should attempt to take complete nutritional and health statuses into account.

CONCLUSIONS
Hypovitaminosis D was common among these case and control subjects, but it was not significantly associated with the presence or severity, as defined by LOS, peripheral oxygen saturation, PICU admission, and PRISA II score, of respiratory illnesses.

REFERENCES