Management of Nutritional Rickets in Indian Children: A Randomized Controlled Trial

by Varun Aggarwal,1 Anju Seth,1 Raman K. Marwaha,2 Bhavna Sharma,2 Pitamber Sonkar,3 Satveer Singh,2 and Satinder Aneja1

1Department of Pediatrics, Kalawati Saran Children’s Hospital and Lady Hardinge Medical College, New Delhi, India
2Department of Endocrinology, Institute of Nuclear Medicine and Allied Science, New Delhi, India
3Department Radiodiagnosis, Kalawati Saran Children’s Hospital and Lady Hardinge Medical College, New Delhi, India

Correspondence: Anju Seth, Department of Pediatrics, Kalawati Saran Children’s Hospital and Lady Hardinge Medical College, New Delhi, India. Tel: 09868206390. E-mail <anjuseth.peds@gmail.com>.

Summary

Introduction: Rickets is usually attributed to vitamin D deficiency. However, recent studies have implicated dietary calcium deficiency in its etiology. Information on relative efficacy of calcium, vitamin D or both together in healing of rickets is limited.

Objective: To study effect of treatment with calcium, vitamin D or a combination of these two on healing of nutritional rickets in young children.

Design: Randomized controlled trial.

Methods: Sixty-seven cases of nutritional rickets in the age group of 6 months to 5 years were randomly allocated to receive vitamin D (600 000 IU single intramuscular dose), calcium (75 mg/kg/day elemental calcium orally) or a combination of the above two for a period of 12 weeks. The demographic parameters, nutritional status, dietary calcium and phytate intake were assessed for all. Radiographs (wrist and knee) and biochemical parameters (serum calcium, inorganic phosphate, alkaline phosphatase, 25-hydroxycholecalciferol and parathyroid hormone) were evaluated at baseline, 6 and 12 weeks for evidence of healing.

Results: Mean dietary intake of calcium in all cases was low (204 ± 129 mg/day). Mean serum 25-hydroxycholecalciferol D level was 15.9 ± 12.4 ng/ml, and 82.1% of patients had serum vitamin D levels < 20 ng/ml, indicative of vitamin D deficiency. After 6 and 12 weeks of treatment, radiological and biochemical evidence of healing rickets was observed in all treatment groups, albeit to a variable extent. The combined end point of normal serum alkaline phosphatase and complete radiological healing at 12 weeks was observed in 50% subjects on combination therapy as compared with 15.7% subjects on vitamin D alone and 11.7% on calcium alone.

Conclusions: Children with rickets had a low serum vitamin D level and a low dietary calcium intake. The best therapeutic response was seen with a combination of vitamin D and calcium than either of them given alone.

Trial registration number: CTRI/2010/091/000448

Key words: nutritional rickets, vitamin D, calcium.

Introduction

Rickets, a common nutritional disorder, is usually attributed to vitamin D deficiency. However, in the past few decades, studies from some tropical countries have shown that calcium deficiency may play a more important role in the causation of rickets [1–6]. In Nigerian children with rickets [1], calcium supplementation alone was as effective as a combination of vitamin D and calcium in inducing healing. Studies among both adult [7] and children [8] in India have shown that calcium intake of our population is much below the recommended allowance. The diet in India is predominantly vegetarian, based on cereals and legumes, and is often deficient in milk and milk products [9]. The low calcium content of the diet is further compromised by the high level of phytates in the vegetarian food. Also, studies from many nations across the globe, including India, have shown a wide prevalence of vitamin D deficiency [10–13]. In a recent study, Seth et al. [14] found serum 25-hydroxycholecalciferol D (25OHD) < 10 ng/ml in 47.8% of apparently healthy lactating mothers and 43.2% of their young infants. Because both calcium and vitamin D deficiencies are likely to be present in children in India and other Asian countries, the relative contribution of each in the etiology of rickets and...
optimum therapeutic strategy for its treatment is not defined. We endeavored to study the effect of supplementation with calcium, vitamin D or a combination of these two on healing of nutritional rickets in Indian children.

**Subjects and Methods**

**Subjects**
We recruited 100 consecutive children aged 6 months to 5 years with clinical and radiological features of nutritional rickets from the outpatient department of a tertiary care teaching hospital in North India from December 2007 to January 2009. Patients with features suggestive of non-nutritional etiology (renal or hepatic disease, malabsorption states, anti-epileptic drug intake or any chronic illness) were excluded from the study, as were cases presenting with hypocalcemic seizures or with a history of consuming calcium or vitamin D supplements in the preceding 6 months. Relevant laboratory evaluation was carried out to exclude non-nutritional rickets where indicated. Participation was voluntary, and informed written consent was taken from the parent or guardian of each child. The study protocol was approved by the Institutional ethical committee.

Of the 100 children screened, 18 had taken calcium/vitamin D supplements in the preceding 6 months, 5 had a chronic medical/surgical disease, 7 were outside the eligible age range and 3 refused consent. Hence, a total of 67 cases were included for the study (Fig. 1).

**Data and sample collection**
The recruited subjects were evaluated for demographic parameters (age, sex, and monthly family income). A detailed dietary evaluation, including history of breast feeding and complementary feeding, was obtained. Dietary assessment was done using 24-h dietary recall and food frequency questionnaire to calculate the dairy consumption of calcium, phosphates and phytates. Information was also sought about the nature, onset and duration of the presenting features. Height (length for children <2 years of age) and weight were recorded, and Z-scores for weight for age and height for age were calculated using World Health Organization 2006 growth reference standards [15].

**Biochemical measurements**
Biochemical evaluation included serum calcium, phosphate, alkaline phosphatase (ALP), 25OHD and parathyroid hormone (PTH) levels. Serum calcium was measured by calorimetric method [normal range for calcium (total) 8.8–10.8 mg/dl, with an analytical sensitivity of 0.2 mg/dl, and calcium (ionic) 4.4–5.4 mg/dl]. Serum phosphate and ALP were determined by photometric analysis (normal range for serum phosphate 3.8–6.5 mg/dl, analytical sensitivity 0.3 mg/dl and of serum ALP 420 IU/l, with analytical sensitivity of 5 IU/l).

Serum 25OHD and PTH levels were measured by electrochemiluminescence assay using a Cobas kit. The analytical sensitivity of the assay was 4 ng/ml.

**Fig. 1.** Study flow chart.
The normal range for serum PTH was 15–65 pg/ml, with analytical sensitivity of 1.2 pg/ml. Based on currently accepted pediatric standards, serum 25OHD levels of <20 ng/ml were defined as vitamin D deficiency [16,17].

Radiological evaluation
Radiographs of left wrist and knee were obtained and were evaluated by two separate observers using the method developed by Thacher et al. [18] on a 0–10-point scale. Mean value of the two scores was used for the analysis. The interclass correlation of scores observed between observers was 0.90, and the correlation within observers was ≥0.91, indicating good reproducibility. A radiological score of >1.5 indicated rickets.

Randomization and treatment allocation
The subjects were randomized using block randomization to one of the following three treatment arms:

- **Group 1**: 600 000 IU vitamin D single intramuscular injection.
- **Group 2**: 600 000 IU vitamin D single intramuscular injection and 75 mg/kg elemental calcium in three divided doses per day for 12 weeks.
- **Group 3**: 75 mg/kg elemental calcium in three divided doses per day for 12 weeks.

Allocation concealment was achieved using opaque sealed envelopes. The subjects were reassessed 6 and 12 weeks after initiation of therapy for evidence of healing by repeat evaluation of the radiological score and measurement of serum calcium (total and ionic), inorganic phosphate and ALP. Serum 25OHD and serum PTH were reassessed at 12 weeks. Both the radiologist and the biochemist were blinded to treatment protocol.

Statistical analysis
The primary variables for comparison between the three treatment groups were improvement in radiological score and biochemical parameters of healing of rickets at 12 weeks. The data were analyzed using Windows SPSS software (version 10). Parametric data were reported as mean ± SD. Non-parametric data were reported as median with interquartile range. The parametric variables were compared using the Student’s *t*-test and non-parametric variables by the Mann-Whitney *U*-test. Dichotomous variables were compared using the *χ*² test. Pearson and Spearman correlation coefficients were used to find correlation between parametric and non-parametric variables, respectively.

**Results**
Of the cases, 20 presented with complaints ascribable primarily to rickets (bow legs 14, delayed walking 14). In the remaining 47, rickets was picked as an incidental finding. In these children, the presenting illnesses were lower respiratory tract infection (37), upper respiratory tract infection (6) and acute gastroenteritis (2). Two patients were siblings of other patients called during subsequent visits.

Baseline characteristics of the cases are shown in Table 1. The groups were comparable for all parameters except for the difference in the serum calcium (ionic) between groups 2 and 3, dietary phosphate intake between groups 1 and 2 and dietary fiber and oxalates between groups 1 and 3. Forty-one children were breast-fed at presentation (including 30 who were exclusively breast-fed). Because it was not feasible to measure the milk output of their mothers, we evaluated the diet for various constituents only for those children (n = 26) who were not breast-fed at presentation.

Figure 2 shows the frequency distribution of serum 25OHD among the study subjects. In all, 82.1% (55/67) subjects had serum 25OHD <20 ng/ml, indicative of vitamin D deficiency. A significant negative correlation (*r* = −0.279) was observed between dietary calcium intake and radiological score (*p* = 0.0287). The correlation between dietary calcium intake and serum PTH was also significant (*r* = −0.26; *p* = 0.02). There was no correlation between dietary calcium intake and serum ALP and inorganic phosphate or between serum 25OHD and radiological score.

Fifty-six patients (19, 20 and 17 in groups 1, 2 and 3, respectively) were followed up until 12 weeks (Fig. 1). All cases showed radiological and biochemical evidence of healing at 6 and 12 weeks of treatment, irrespective of the treatment arm. However, the improvement was to varying degrees (Tables 2 and 3 and Fig. 3). The best radiological response was observed in group 2, in which 70% of cases had a mean radiological score ≤1.5, indicative of complete healing at 12 weeks, as opposed to 42.1% in group 1 and 23.5% in group 3. Likewise, 70% of subjects in group 2 as opposed to 36.8% in group 1 and 17.6% in group 3 achieved normalization of serum ALP. The combined end point of a normal serum ALP and radiological evidence of complete healing at 12 weeks was seen in 50% of patients in group 2 as compared with 15.7% in group 1 and 11.7% in group 3.

It was noteworthy that even with such a large dose of vitamin D, 15 (78.9%) children of group 1 and 11 (55%) children of group 2 still had serum 25OHD <20 ng/ml at 12 weeks. The mean sera 25OHD in groups 1 and 2 were 32.8 ± 23.8 and 31 ± 31 ng/ml, respectively.

**Discussion**
Nutritional rickets is usually thought to be caused by deficiency of vitamin D. However, studies from
tropical countries (Nigeria [1] and South Africa [19]) have shown that low intake of calcium in the diet might be more important in the causation of rickets. Dietary calcium intake of the Indian population is very low [7,8]. Also vitamin D deficiency is common in Indian children [13,14]. In this study, we have explored the relative efficacy of calcium and vitamin D given alone or in combination in treatment of nutritional rickets. Several studies [1–5] have shown that calcium supplementation with or without vitamin D therapy is more effective than supplementation with vitamin D alone in inducing healing in rickets. Our results show that combined therapy with calcium and vitamin D produces better healing in nutritional rickets as compared with either given alone.

We found that 82.1% subjects had serum 25OHD levels <20 ng/ml, indicative of vitamin D deficiency. Several other studies [2,6] have reported similar serum vitamin D levels in cases of rickets. Thus, vitamin D deficiency is invariable in cases of rickets. We also found very low dietary calcium intake in our patients (204 ± 129 mg/day), much lower than the Indian Council of Medical Research (ICMR)-[8] and Institute of Medicine (IOM)-[20] recommended daily allowance of 500 mg/day and 700–1000 mg/day, respectively. Low dietary intake of calcium in

### Table 1

Baseline characteristics of study subjects in the three groups

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group 1 (n = 23)</th>
<th>Group 2 (n = 22)</th>
<th>Group 3 (n = 22)</th>
<th>P (1 vs. 2)</th>
<th>P (2 vs. 3)</th>
<th>P (3 vs. 1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (months)</td>
<td>16.48 ± 11.96</td>
<td>17.05 ± 10.67</td>
<td>19.27 ± 13.04</td>
<td>0.98</td>
<td>0.54</td>
<td>0.46</td>
</tr>
<tr>
<td>Age range (months)</td>
<td>6–58</td>
<td>6–60</td>
<td>6–60</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Gender</td>
<td>M = 13, F = 10</td>
<td>M = 9, F = 13</td>
<td>M = 13, F = 9</td>
<td>—</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Height Z-score</td>
<td>−1.72 ± 1.00</td>
<td>−1.59 ± 1.06</td>
<td>−1.82 ± 1.17</td>
<td>0.66</td>
<td>0.49</td>
<td>0.75</td>
</tr>
<tr>
<td>Weight Z-score</td>
<td>−1.39 ± 1.10</td>
<td>−1.77 ± 0.96</td>
<td>−1.34 ± 1.12</td>
<td>0.23</td>
<td>0.18</td>
<td>0.87</td>
</tr>
<tr>
<td>UV score (min m²/d)</td>
<td>1.78 ± 2.82</td>
<td>1.96 ± 3.0</td>
<td>1.74 ± 2.70</td>
<td>0.83</td>
<td>0.83</td>
<td>0.96</td>
</tr>
<tr>
<td>Dietary total calcium intake (mg/d)</td>
<td>204.0 ± 182.67</td>
<td>202.25 ± 91.66</td>
<td>207.4 ± 100.2</td>
<td>0.98</td>
<td>0.91</td>
<td>0.89</td>
</tr>
<tr>
<td>Dietary dairy calcium intake (mg/d)</td>
<td>88.70 ± 156.02</td>
<td>73.55 ± 112.77</td>
<td>94.27 ± 124.4</td>
<td>0.98</td>
<td>0.57</td>
<td>0.95</td>
</tr>
<tr>
<td>Dietary fiber intake (mg/d)</td>
<td>0.41 ± 0.30</td>
<td>0.55 ± 0.54</td>
<td>1.1 ± 0.67</td>
<td>0.49</td>
<td>0.08</td>
<td>0.01*</td>
</tr>
<tr>
<td>Dietary phosphate (mg/d)</td>
<td>274.0 ± 132.2</td>
<td>490.29 ± 249.45</td>
<td>360.97 ± 86.67</td>
<td>0.03*</td>
<td>0.14</td>
<td>0.10</td>
</tr>
<tr>
<td>Dietary phytates (mg/d)</td>
<td>29.83 ± 25.0</td>
<td>16.55 ± 17.93</td>
<td>26.57 ± 21.90</td>
<td>0.23</td>
<td>0.312</td>
<td>0.75</td>
</tr>
<tr>
<td>Dietary oxalates intake (mg/d)</td>
<td>4.7 ± 3.94</td>
<td>7.84 ± 5.08</td>
<td>7.8 ± 2.4</td>
<td>0.16</td>
<td>0.16</td>
<td>0.04*</td>
</tr>
</tbody>
</table>

*p < 0.05.

Fig. 2. Frequency distribution of serum 25OHD levels in study subjects (n = 67) at presentation.
children with rickets has also been reported by Balasubramanian et al. [6] from India and Thacher et al. [2] from Nigeria. Regression analysis showed significant correlation of radiological score with dietary calcium intake but not with serum 25OHD levels. It is likely that the existing vitamin D deficiency in the subjects is exaggerated by hyperparathyroidism induced by calcium deficiency. This phenomenon has been reported by Clements et al. [21], who have shown in animal studies that the rate of inactivation of vitamin D in the liver is increased by calcium deprivation. The effect is mediated by 1,25-dihydroxyvitamin D produced in response to secondary hyperparathyroidism, which promotes the hepatic conversion of vitamin D to inactivated polar products. Low vitamin D status, in turn, impairs calcium absorption from the intestine, thereby exacerbating calcium deficiency. Thus, a combination of low dietary calcium and suboptimal vitamin D status will enhance both deficiencies and precipitate development of rickets in children. The main source of calcium in the study population was milk and other dairy products. Phytates, phosphates and fiber in the diet was predominantly from the cereals and vegetables. Wheat is staple food in the study population and was found to be the predominant contributor for the phytates and fiber.

We compared three different treatment modalities for their efficacy in healing rickets. Although healing was observed in all the three treatment groups, earlier and better healing, as determined by the combined end point of a normal serum ALP, and complete radiological healing were observed when both vitamin D and calcium were given together than with administration of either alone. Because only half of the patients in group 2, the group with the best

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Group 1 (%)</th>
<th>Group 2 (%)</th>
<th>Group 3 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum ALP (IU/l)</td>
<td>34.5 ± 13.8</td>
<td>42.9 ± 12.3</td>
<td>34.2 ± 0.2</td>
</tr>
<tr>
<td>Serum PTH (pg/ml)</td>
<td>69.0 ± 18.1</td>
<td>65.6 ± 28.8</td>
<td>49.7 ± 46.0</td>
</tr>
<tr>
<td>Radiological score</td>
<td>70.4 ± 9.3</td>
<td>82.2 ± 5.8</td>
<td>59.6 ± 15.8</td>
</tr>
</tbody>
</table>
response, showed complete evidence of healing in terms of a normal radiological score and a normal serum ALP; results of this study also indicate that continued treatment is required beyond 12 weeks to ensure complete healing.

Like our observation, Kutluk et al. [22] from Italy found supplementation with both vitamin D and calcium produced a better response in healing nutritional rickets as compared with calcium or vitamin D given alone. However, Thacher et al. [1] from Nigeria found that treatment with calcium alone induced healing similar to a calcium and vitamin D combination, whereas a smaller proportion of subjects on vitamin D alone manifested healing. A previous study on nutritional rickets from India [6] has reported complete biochemical and radiological healing of rickets at the end of 3 months in patients receiving calcium alone or calcium with vitamin D. However, a poor follow-up rate (40%) and the use of variable doses of vitamin D in the group receiving combination therapy (6000 IU of oral vitamin D daily for 3 months for some patients and a 600 000-IU single oral dose for some patients) in that study limit the interpretation of its results.

There are some concerns about megadose therapy leading to hypercalcemia and hypercalciuria. We measured spot non-fasting urine calcium/creatinine ratio in all patients, irrespective of the treatment arm. Two patients who received a combination of calcium and vitamin D developed asymptomatic hypercalcemia and hypercalciuria at 3 and 4 weeks after initiation of therapy. However, these patients were then followed weekly with monitoring of serum calcium and urine calcium/creatinine ratio. The hypercalcemia and hypercalciuria resolved within the next 4 weeks in both the patients. None of the two demonstrated any evidence of renal calcification on ultrasonologic evaluation at onset, 2, 3 and 6 months after initiation of therapy. However, studying toxicity was not the primary objective of the study, and the numbers were not sufficient to draw any conclusions for safety issues.

Our study has a few limitations that we would like to acknowledge. The sample size was small, and the...
duration of follow-up was only 12 weeks, during which complete healing was not achieved in most cases. Nevertheless, the results indicate that children with rickets have vitamin D deficiency and a low dietary intake of calcium. Best therapeutic results are obtained when these patients are treated with both vitamin D and calcium. Supplementation with vitamin D and calcium needs to continue beyond 12 weeks to ensure complete healing.

References