Iron supplementation improves iron status and reduces morbidity in children with or without upper respiratory tract infections: a randomized controlled study in Colombo, Sri Lanka

Angela de Silva, Sunethra Atukorala, Irangani Weerasinghe, and Namanjeet Ahluwalia

ABSTRACT

Background: Iron deficiency anemia and recurrent infections are common among children of low socioeconomic status.

Objective: The objective was to evaluate the effects of iron supplementation on iron status and morbidity in children with or without infection.

Design: Children aged 5–10 y were recruited for a randomized, controlled, double-blind study from outpatients attending the Children’s Hospital, Colombo, Sri Lanka. Clinical, inflammatory, nutritional, and iron statuses were determined at baseline and after the intervention. Children with a history of recurrent upper respiratory tract infections (URTIs) and with laboratory and clinical evidence of a current URTI constituted the infection group \((n = 179)\), and children without infection constituted the control group \((n = 184)\). Subjects in both groups were supplemented with ferrous sulfate \((60 \text{ mg Fe})\) or placebo once daily for 8 wk. Morbidity from URTIs, the number of gastrointestinal infections, and compliance were recorded every 2 wk.

Results: The overall prevalence of anemia was 52.6\%. Iron supplementation significantly improved iron status by increasing hemoglobin \((P < 0.001)\) and serum ferritin \((P < 0.001)\) concentrations from baseline values in the children with or without infection. There was no significant improvement in iron status in the children who received placebo. In both the infection group and the control group, the mean number of URTI episodes and the total number of days sick with URTI during the period of intervention were significantly lower \((P < 0.005\) and \(P < 0.001,\) respectively) in the children who received iron supplements than in those who received placebo.

Conclusion: Iron supplementation significantly improves iron status and reduces morbidity from URTIs in children with or without infection.

KEY WORDS Anemia, iron status, inflammatory indicators, upper respiratory tract infections, iron supplementation, 5–10-y-old children, Sri Lanka

INTRODUCTION

Iron deficiency anemia is a major public health problem in developing countries including Sri Lanka. A National Health and Nutrition Survey in 1995 found that 58\% of primary school children were anemic when a hemoglobin concentration cutoff of < 12 \text{ g/dL} was used \((1)\). The prevalence of anemia was found to be especially high among children from low socioeconomic groups; such children live in crowded environments and are also prone to recurrent infections. Iron deficiency is the major cause of anemia in Sri Lanka (Ministry of Health, Sri Lanka, Policy Document 2000). Prevention of iron deficiency is essential because previous studies highlighted the adverse effects of iron deficiency on cognitive development, attention, behavior, school performance, and physical activity in children \((2–5)\). Furthermore, iron deficiency is also associated with impaired immunocompetence and therefore can lead to increased morbidity \((6–8)\).

The relation between iron status and morbidity is controversial \((9)\). Some longitudinal studies with oral iron therapy showed a reduced prevalence of respiratory and gastrointestinal infections \((10–12)\). Other reports indicated that there was an increase or no change in the incidence of infectious disease \((13–15)\). Difficulties in interpreting the conflicting results from these studies on iron therapy and infection include limitations in experimental design, such as the inclusion of severely malnourished subjects; lack of proper controls; and the presence of other confounding variables, such as other nutritional deficiencies \((\text{eg, of protein, folic acid, or vitamin B-12})\). Furthermore, many of the above studies were carried out in countries or areas where malaria is endemic and becomes a complicating factor \((15, 16)\). In addition, most studies on children were limited to infants or preschoolers. Hence, there is a need for carefully designed and executed studies to examine the effects of iron supplementation on morbidity in primary school children of low socioeconomic status, because recurrent respiratory and gastrointestinal infections affect their well-being and lead to irregular school attendance. It is possible that iron supplementation could improve iron status and reduce morbidity from upper respiratory tract infections (URTIs) and gastrointestinal infections in children of this age group. Therefore, while taking into account

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possible confounding factors, we carried out a study to evaluate the effect of iron supplementation (iron compared with placebo) on iron status and morbidity from URTIs and gastrointestinal infections in 5–10-y-old children with or without infection.

SUBJECTS AND METHODS

Subjects

Four hundred fifty-three children (251 boys and 202 girls) who were with or without infection and were aged between 5 and 10 y were recruited over a period of 2 y. The subjects were children seeking treatment at the Out Patients Department of the Lady Ridgeway Children’s Hospital, Colombo, Sri Lanka, which is a non-fee-levying, state-run hospital.

The study was approved by the Ethical Review Committee of the Faculty of Medicine, University of Colombo, Sri Lanka, and by The Pennsylvania State University, University Park. Children were recruited into the study after obtaining written informed consent from their parents or guardians. Children with severe anemia at baseline were excluded from the study, supplemented with iron, and followed up. All subjects (iron and placebo groups) who were anemic at the end of the study were supplemented with iron.

Inclusion criteria

The infection group consisted of children with a past history of recurrent URTIs and clinical and laboratory evidence of a current URTI. The operational definition of recurrent URTIs was ≥2 separate episodes/mo over the preceding 6–12 mo (17). URTIs included coryza, rhinitis, acute sinusitis, pharyngitis, tonsillitis, laryngitis, and tracheitis (18). The symptoms elicited were runny nose, cough, sore throat, difficulty in breathing or wheeze, and earache or discharge, with or without fever. Laboratory evidence of infection consisted of ≥3 of the following indicators of inflammation: an erythrocyte sedimentation rate (ESR) >20 mm/h, an α1-acid glycoprotein concentration >1.1 g/L, a C-reactive protein concentration >10 mg/L, and a white blood cell (WBC) count >9 × 10³/mm³ (19–21). Episodes of illness were considered separate if there were ≥5 symptom-free days between the episodes. The group without infection (ie, the control group) consisted of children seeking treatment for minor injuries that did not involve infection or for poor vision. These children either had no elevated values for any of the 4 laboratory tests of inflammation or had an elevated value for only 1 of the tests.

Exclusion criteria

Children with severe protein-energy malnutrition, a history of other systemic diseases, chronic bronchial asthma, or chronic diarrhea were excluded from the study. Children were also excluded if they had signs of an infection but had elevated values for ≤2 inflammatory indicators, if hemoglobin data was not available, or if the children had a hemoglobin concentration <70 g/L, which indicated severe anemia at baseline (22).

Methods

The study was a longitudinal, randomized, controlled, double-blind supplementation trial, and the study design is shown in Figure 1. On the basis of the estimated prevalence of anemia in this age group in Sri Lanka (1), the sample size was calculated to be 420, with the assumption that the prevalence of anemia in children with infection was similar to that in children without infection and with the assumption of a 20% dropout rate.

Baseline (before supplementation)

For each child, a detailed medical history was obtained and a clinical examination was carried out by the study pediatrician. Socioeconomic status and morbidity from respiratory and gastrointestinal illnesses (> 3 loose stools/24 h) during the preceding 2 wk were recorded on pretested, interviewer-administered questionnaires. Height to the nearest 0.5 cm and weight to an accuracy of 0.5 kg (Salter Balance; Salter Scales, London) were recorded. A venous blood sample (5 mL) was obtained from each child between 0900 and 1100 by using sterile equipment. An aliquot (2.5 mL) was collected in tubes containing an anticoagulant (EDTA), placed on ice, and processed within 3 h of collection. The remaining blood (2.5 mL) was collected in a tube without anticoagulants, serum was separated by centrifugation at 3000 rpm (MSE Soniprep 150; MSE Scientific Instruments, Crawley, Sussex, United Kingdom) for 7 min at 28°C, and aliquots were stored at −20°C. The lag period between baseline measurements and the start of the intervention was 24–36 h.

Supplementation and follow-up

Children were stratified by age and sex and were randomly assigned to receive iron or placebo on a 3:1 basis within each stratum. Because variation in the response to the intervention was likely to be greater in the iron group than in the placebo...
group, 3 times as many subjects were assigned to the iron group as were assigned to the placebo group. The allocation of more children to the iron group ensured an adequate number in the intervention group to examine the effects of the intervention. One child with severe anemia was treated with iron and followed up separately.

The intervention consisted of iron (ferrous sulfate) tablets (Feosol; Smith Kline Beecham Inc, Philadelphia) containing the recommended daily dose of 60 mg elemental Fe (23) or placebo capsules containing 0.5 g lactose (State Pharmaceuticals Corporation, Colombo, Sri Lanka). The parents or guardians of each child were instructed to give the child one tablet/d at night. Neither the subjects nor the members of the field staff were aware of the nature of the supplement. During the follow-up period, field investigators visited the homes of the participants every 2 wk to monitor compliance by questioning participants and counting unused tablets. The parents or caregivers were requested to make note of morbidity from URTIs and gastrointestinal infections for the preceding 2 wk. Field investigators verified these data during home visits by questioning the parents or guardians with the use of interviewer-administered questionnaires.

**After the intervention**

After 8 wk of supplementation, the children were reassessed at the hospital. All assessments made before the intervention were repeated, and morbidity and compliance data were recorded. Subjects in both the iron and placebo groups who were anemic [hemoglobin concentration < 115 g/L (23)] after the intervention were supplemented with iron (60 mg elemental Fe/d) for 4 wk and referred to the Out Patients Department for follow-up.

**Laboratory methods**

A complete blood count including hemoglobin, hematocrit, mean cell volume, and WBC count was obtained by using an automated analyzer (Cel-dyn 3500; Abbott Diagnostics, Abbott Park, IL). ESR was measured by using a modified version of the Westergren method (Dispette 2; Ulster Medical Products, a division of Lukens Medical Corporation, Albuquerque, NM), serum C-reactive protein concentrations were measured by using latex agglutination (bioMérieux sa, Marcy-l’Etoile, France), and α₁-acid glycoprotein concentrations were measured by using radial immunodiffusion (Kent Laboratories Inc, Redmond, WA). Iron stores were assessed by serum ferritin determination (immunoradiometric assay; DPC Inc, Los Angeles). Baseline concentrations of potential confounders, serum folic acid, and vitamin B-12 (SimulTRAC SNB Radioassay; ICN Pharmaceuticals, Orangeburg, NY) were also measured. All samples were assayed in duplicate, and a pooled serum sample was used as an internal control.

**Statistical analysis**

Completed questionnaires were checked before the data were entered into an IBM PC 300 GL, and the data were analyzed by using SPSS version 7.5 for WINDOWS (SPSS Inc, Chicago) and EPI-INFO version 6.04 (Centers for Disease Control and Prevention, Atlanta). The baseline characteristics of the children with or without infection were examined by chi-square test and analysis of variance. Serum ferritin data were log transformed before statistical analysis because they had a skewed distribution. Changes in iron status (hemoglobin and serum ferritin) and inflammatory indicators (ESR, α₁-acid glycoprotein, and WBC) after the intervention were analyzed by using a two-factor analysis of variance that included treatment and infection main effects and a treatment × infection interaction effect. The effects of supplementation (iron or placebo) on morbidity from URTIs and gastrointestinal infections were examined by using a two-factor analysis of variance that included treatment and infection main effects and a treatment × infection interaction effect. If interactions were significant, the data were analyzed separately for children with or without infection (24). Statistical significance was set at \( P < 0.05 \).

**RESULTS**

**Baseline**

Three hundred sixty-three children completed the full course of supplementation and follow-up, and 71 children dropped out of the study. Twelve children who had infections but had elevated values for only 2 inflammatory indicators at baseline, 6 children who lacked baseline hemoglobin data, and 1 child with a baseline hemoglobin concentration < 70 g/L did not fulfill the inclusion criteria for the study. Subjects dropped out of the study because their families were displaced due to floods (\( n = 26; 36.6\% \)) or migrated to other areas (\( n = 20; 28.1\% \)) or because the subjects failed to show up at the hospital or provide a blood sample after the intervention (\( n = 25; 35.2\% \)). The children who dropped out of the study were not significantly different from the remaining children in age, body mass index (BMI; in kg/m²), or baseline hemoglobin concentration. The mean ± SD age of the children in the study was 6.7 ± 1.4 y (Table 1). Most of the children belonged to low socioeconomic groups, and there were no significant differences between the iron and placebo groups in socioeconomic indicators, such as mother’s education level and family income (Table 1) and housing and latrine facilities (data not shown).

Mean BMI values in the children with infection did not differ significantly between the iron and placebo groups (Table 1). In the children without infection, those who were supplemented with iron had a significantly lower BMI (\( P = 0.007 \)) than did those who were supplemented with placebo. None of the children had severe protein-energy malnutrition as indicated by BMI values below the 5th percentile (25). Serum folic acid and vitamin B-12 concentrations did not differ significantly between the children with or without infection or between the iron and placebo groups. Low serum folic acid concentrations (< 3 ng/mL (26)) were observed in 20% and 24.8% of the children with and without infection, respectively, and low serum vitamin B-12 concentrations (< 150 pg/mL (26)) were observed in 0.6% and 0.7% of the children with and without infection, respectively.

**Compliance and side effects**

A high compliance was noted in both the iron groups (with infection: 95.1%; without infection: 95.5%) and the placebo groups (with infection: 97.3%; without infection: 96.9%). Only a few children experienced side effects in either the iron group (constipation: 11.7%; vomiting: 1.6%) or the placebo group (constipation: 1%; vomiting: 1.1%; skin rash: 1.9%).

**Effects of iron supplementation on inflammatory indicators, iron status, and morbidity**

When tests of inflammation were considered at baseline, the children with infection had significantly higher ESRs, α₁-acid...
The overall prevalence of anemia (hemoglobin concentration <115 g/L) in the study population was 52.6% at baseline, and there was no significant difference in the prevalence of anemia between the children with or without infection or between the iron and placebo groups (Table 3). Children with infection had significantly higher (P < 0.001) serum ferritin concentrations than did those without infection. Serum ferritin concentrations did not differ significantly between the iron and placebo groups either in the children with infection or in those without infection (Table 3). After supplementation, the prevalence of anemia in the iron groups was significantly lower than that at baseline. The effect of supplementation was examined by comparing the changes in hemoglobin and ferritin concentrations after supplementation in the iron and placebo groups. Iron supplementation had a significant (P < 0.001) effect in increasing the mean change in hemoglobin concentration in the children with or without infection. The presence or absence of infection had no significant influence (P = 0.415) on the change in hemoglobin concentration. Serum ferritin also increased significantly from baseline in the children with or without infection who were supplemented with iron. The change in serum ferritin was influenced by iron supplementation (P < 0.001) and the presence or absence of infection (P < 0.005). Infection ×

glycoprotein concentrations, and WBC counts than did those without infection (Table 2). Furthermore, among the children with infection, there were no significant differences in the concentrations of inflammatory indicators between the iron and placebo groups. After the intervention, the values of all inflammatory indicators except WBC significantly decreased (P < 0.05) in the children with infection but not in the children without infection. However, the effect of treatment (iron compared with placebo) on the change in inflammatory indicators was not significant.

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Table 1
Baseline characteristics of the study population

<table>
<thead>
<tr>
<th>Socioeconomic indicators (%)</th>
<th>Children with infection (n = 179)</th>
<th>Children without infection (n = 184)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Iron group (n = 127)</td>
<td>Placebo group (n = 52)</td>
</tr>
<tr>
<td>Age (y)</td>
<td>6.5 ± 1.3</td>
<td>6.7 ± 1.3</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>12.9 ± 2.1</td>
<td>12.9 ± 1.2</td>
</tr>
<tr>
<td>Folic acid (ng/mL)</td>
<td>4.91 ± 2.7</td>
<td>5.79 ± 3.6</td>
</tr>
<tr>
<td>Vitamin B-12 (pg/mL)</td>
<td>622 ± 209</td>
<td>629 ± 232</td>
</tr>
</tbody>
</table>

Socioeconomic indicators (%)

<table>
<thead>
<tr>
<th>Family income</th>
<th>Children with infection (n = 179)</th>
<th>Children without infection (n = 184)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;3500 rupees/mo (&lt;$40)</td>
<td>44.1 [56]</td>
<td>44.2 [23]</td>
</tr>
<tr>
<td>≥3500 rupees/mo (≥$40)</td>
<td>55.1 [70]</td>
<td>55.8 [29]</td>
</tr>
</tbody>
</table>

Mother’s educational level

| Primary or secondary         | 66.0 [82]                         | 67.3 [35]                           | 66.2 [86]           | 64.6 [31]              |
| Tertiary                     | 29.9 [38]                         | 25.0 [13]                           | 29.2 [38]           | 27.1 [13]              |

1 Significantly different from the iron group, P < 0.01.
2 n in brackets. Socioeconomic data were unavailable for 1 child with infection and 6 children without infection (iron group: n = 4; placebo group: n = 2).

Table 2
Baseline values of and changes in inflammatory indicators after supplementation with iron or placebo

<table>
<thead>
<tr>
<th>Group</th>
<th>ESR²</th>
<th>AGP¹</th>
<th>WBC³</th>
<th>CRP &gt; 10 mg/L³</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>Change</td>
<td>Baseline</td>
<td>Change</td>
</tr>
<tr>
<td>Children with infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron group (n = 127)</td>
<td>31.6 ± 15.2</td>
<td>-15.4 ± 1.0</td>
<td>1.42 ± 0.3</td>
<td>-0.44 ± 0.3</td>
</tr>
<tr>
<td>Placebo group (n = 52)</td>
<td>30.3 ± 14.3</td>
<td>-12.9 ± 1.5</td>
<td>1.41 ± 0.4</td>
<td>0.08 ± 0.5</td>
</tr>
<tr>
<td>Children without infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron (n = 134)</td>
<td>11.8 ± 4.5</td>
<td>0.029 ± 0.9</td>
<td>0.78 ± 0.22</td>
<td>-0.44 ± 0.5</td>
</tr>
<tr>
<td>Placebo (n = 50)</td>
<td>11.7 ± 3.5</td>
<td>-0.52 ± 1.5</td>
<td>0.68 ± 0.20</td>
<td>0.03 ± 0.3</td>
</tr>
</tbody>
</table>

1 Except for CRP, all baseline values are means ± SDs. All change values are means ± SEs. ESR, erythrocyte sedimentation rate; AGP, α1-acid glycoprotein; WBC, white blood cell; CRP, C-reactive protein. All baseline values in the children with infection were significantly higher (P < 0.005) than those in the children without infection.
2 The effect of infection was significant (P < 0.001), but the effect of supplementation (P = 0.174) and the infection × supplementation interaction effect (P = 0.24) were not significant.
3 The effect of infection was significant (P < 0.001), but the effect of supplementation (P = 0.925) and the infection × supplementation interaction effect (P = 0.354) were not significant.
4 The effect of infection was significant (P < 0.001), but the effect of supplementation (P = 0.441) and the infection × supplementation interaction effect (P = 0.942) were not significant.
5 n in brackets. Mean values were not available for CRP data because the CRP assay was semiquantitative.
6 Significantly different from baseline, P < 0.005.
iron supplementation interactions were not significant for hemoglobin ($P = 0.303$) or serum ferritin ($P = 0.783$) concentrations.

The severity of infection at baseline (defined by the presence of a documented fever accompanying an URTI) was compared between the iron and placebo groups. Fever was defined as an axillary temperature $\geq 37.5$°C (27). At baseline the percentage of children who had URTIs with fever in the iron group (51.5%) was not significantly different from that in the placebo group (51.9%). During follow-up, the iron-supplemented children with infection had a significantly lower ($P < 0.005$) number of URTI episodes and a significantly lower number of URTI episodes accompanied by fever ($P < 0.005$) than did the children who were given placebo; both documented and undocumented episodes of fever were included in this analysis (Table 4). The children without infection who were supplemented with iron also had a significantly lower number of URTI episodes ($P < 0.001$) than those who received placebo.

A significant supplementation $\times$ infection interaction ($P = 0.003$) was noted for duration of illness (Table 4). The children with or without infection who were supplemented with iron had a significantly ($P < 0.001$) lower number of days of illness than did the corresponding placebo groups (Table 4). The number of episodes of gastrointestinal infection was low in all groups. Although the children with infection in the iron group had a significantly higher number of episodes of gastrointestinal infections than did those in the placebo group, the effect of iron supplementation was not

### Table 3
Baseline values of and changes in hemoglobin and serum ferritin concentrations after supplementation with iron or placebo

<table>
<thead>
<tr>
<th>Iron-status indicator</th>
<th>Children with infection ($n = 179$)</th>
<th>Children without infection ($n = 184$)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>Change</td>
</tr>
<tr>
<td><strong>Hemoglobin</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All subjects (g/L)$^1$</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron</td>
<td>112.6 ± 10.4</td>
<td>6.44 ± 0.82</td>
</tr>
<tr>
<td>Placebo</td>
<td>115.8 ± 10.3</td>
<td>0.74 ± 1.05</td>
</tr>
<tr>
<td>$&lt;115$ g/L (%)$^2$</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron</td>
<td>59.1 [75/127]</td>
<td>32.3 [41/127]$^3$</td>
</tr>
<tr>
<td>Placebo</td>
<td>46.2 [24/52]</td>
<td>42.3 [22/52]</td>
</tr>
<tr>
<td><strong>Serum ferritin</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All subjects (μg/L)$^4$</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron</td>
<td>46.1 (23.9–88.9)</td>
<td>6.6 ± 3.9</td>
</tr>
<tr>
<td>Placebo</td>
<td>52.4 (25.9–105.9)</td>
<td>−14.9 ± 10.4</td>
</tr>
<tr>
<td>$&lt;20$ μg/L (%)$^5$</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

$^1$Baseline values are means ± SDs, and change values are means ± SEs. The effect of supplementation was significant ($P < 0.001$), but the effect of infection ($P = 0.415$) and the infection $\times$ supplementation interaction effect ($P = 0.318$) were not significant.

$^2$n anemic/total n in brackets.

$^3$Significantly different from baseline, $P < 0.05$.

$^4$Ferritin data were unavailable for 16 children (with infection: $n = 8$; without infection: $n = 8$).

$^5$Baseline values are geometric means and ranges, and change values are means ± SEs. The effects of infection ($P < 0.005$) and supplementation ($P < 0.001$) were significant, but the supplementation $\times$ infection interaction effect was not significant ($P = 0.783$).

$^6$n with low ferritin/total n in brackets.

### Table 4
Effects of supplementation with iron or placebo on morbidity$

<table>
<thead>
<tr>
<th>Group</th>
<th>Number of URTI episodes$^2$</th>
<th>Number of URTI episodes with fever$^3$</th>
<th>Total number of days sick with an URTI$^4$</th>
<th>Number of episodes of gastrointestinal infection$^4$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Children with infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron group ($n = 127$)</td>
<td>2.14 ± 1.1</td>
<td>0.58 ± 0.79</td>
<td>17.2 ± 11.5</td>
<td>0.19 ± 0.43</td>
</tr>
<tr>
<td>Placebo group ($n = 52$)</td>
<td>2.78 ± 0.9</td>
<td>0.92 ± 0.83</td>
<td>26.9 ± 9.7</td>
<td>0.10 ± 0.30</td>
</tr>
<tr>
<td>Children without infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Iron group ($n = 134$)</td>
<td>0.96 ± 0.70</td>
<td>0.12 ± 0.35</td>
<td>5.2 ± 4.5</td>
<td>0.06 ± 0.28</td>
</tr>
<tr>
<td>Placebo group ($n = 50$)</td>
<td>1.38 ± 0.94</td>
<td>0.24 ± 0.47</td>
<td>8.9 ± 6.9</td>
<td>0.04 ± 0.19</td>
</tr>
</tbody>
</table>

$^2$± SD. URTI, upper respiratory tract infection.

$^3$The effects of infection ($P < 0.001$) and supplementation ($P < 0.001$) were significant, but the infection $\times$ supplementation interaction effect was not significant ($P = 0.318$).

$^4$The effects of infection ($P < 0.001$) and supplementation ($P < 0.005$) were significant, but the infection $\times$ supplementation interaction effect was not significant ($P = 0.125$).

$^5$The effects of infection ($P < 0.001$) and supplementation ($P < 0.001$) were significant, as was the infection $\times$ supplementation interaction effect ($P = 0.003$).

$^6$The effect of supplementation was significant ($P = 0.003$), but the but the effect of supplementation ($P = 0.102$) and the supplementation $\times$ infection interaction effect ($P = 0.419$) were not significant.
significant. Among the children with infection, there was no significant difference between the iron and placebo groups in the percentage of children who were given oral antibiotics at recruitment or during follow-up.

The numbers of episodes of URTI morbidity were compared between iron-deficient and iron-sufficient groups. Subjects were considered iron deficient if their increase in hemoglobin concentration after iron supplementation was ≥ 10 g/L and were considered iron sufficient if their increase in hemoglobin concentration after iron supplementation was < 10 g/L (28). Among the children with infection, there was no significant difference in the mean ± SD number of episodes of URTI morbidity between the iron-deficient subjects (2.17 ± 1.1; n = 45) and the iron-sufficient subjects (2.16 ± 1.1; n = 81). A similar result was found for the children without infection: the number of episodes of URTI morbidity did not differ significantly between the iron-deficient (1.03 ± 0.7; n = 45) and iron-sufficient subjects (1.05 ± 0.8; n = 89).

DISCUSSION

Anemia and upper respiratory tract infections are common problems among primary school children of low socioeconomic status, and a complex relation exists between iron status and infection. Iron deficiency and anemia are associated with impaired immunocompetence and increased morbidity (7, 8), and infections can affect iron metabolism (29). Thus, our interest was to examine the effects of iron supplementation on iron status and morbidity in children with or without infection. The study population was comprised of children from low socioeconomic backgrounds with a high prevalence of anemia (52.7%), similar to the prevalence reported in the national survey (1). In this randomized, controlled, double-blind study, high compliance was noted in both the iron and placebo groups. The delivery of the supplements every 2 wk to the participants’ homes by field staff may have enhanced the motivation of parents to provide supplements. Interestingly, the number of children who experienced side effects was low in both the iron and placebo groups, which may also have contributed to the high compliance. In this supervised trial, iron supplementation was associated not only with an improvement in iron status but also with a reduction in morbidity due to URTIs in children with or without infection.

The magnitudes of the reductions in the prevalence of anemia and of the mean changes in hemoglobin concentration after iron supplementation were similar between the children with and without infection. This indicates that children with recurrent URTIs and those without infection can increase their hemoglobin concentrations to a similar extent, suggesting that iron supplementation may be beneficial for children even during an active URTI. Other studies also showed that hemoglobin concentrations improve with iron supplementation even in the presence of inflammatory conditions (14, 30, 31). The percentage of children with low iron stores, as indicated by a serum ferritin concentration <20 µg/L (32), was only 8.7% in the infection group at baseline. This value probably underestimates the proportion with low body iron stores because ferritin is an acute-phase protein that increases during infections (33, 34). The significant increase in serum ferritin concentration that was observed in the iron-supplemented children with or without infection was to be expected in this community with a high prevalence of iron deficiency. To our knowledge, the present study is one of the few to provide important information on changes in serum ferritin concentration after iron supplementation in children with infection.

Iron supplementation had no significant effect on changes in inflammatory indicators. This was probably because the magnitude of the change in concentrations of these indicators was not large enough. The record of incidence of morbidity is a more important and direct indicator of immune function in vivo. Interestingly, in this cohort of school-aged children with or without infection, iron supplementation was associated with reduced morbidity.

Previous studies on iron supplementation during infections yielded conflicting results, with few studies showing beneficial effects of iron on the prevalence of infections (10–12). A lower incidence of respiratory infections was reported in term infants who were fed an iron-containing milk formula than in those who were fed an unfortified formula (11). The positive effect of iron supplementation on linear growth was attributed to decreased morbidity in preschool children in 2 studies (12, 35). In some studies, iron supplementation did not change the incidence of morbidity (14, 36, 37). In comparison with morbidity in a control group, the morbidity of Pakistani infants who were given an iron-fortified cereal during weaning did not change, but a high incidence of malnutrition was a confounding factor in these subjects (14). Heresi et al (37) found that morbidity from respiratory infections and diarrhea did not increase in infants after they were given iron-fortified milk. Furthermore, in another study in Bangladesh, the incidence of diarrhea or respiratory illness in preschoolers did not change after supplementation with iron for 1 y (36).

In contrast, a study of adult Somali nomads who were supplemented with iron showed an increased frequency of infections (13). However, this study lacked adequate controls, and the iron supplement dose that was provided (900 mg FeSO4/d) was much greater than that presently recommended (23). In the present study, infection and supplementation were both significant main effects in all aspects of morbidity that were examined. The children with infection had a greater number of URTI episodes, more severe episodes, and episodes of a longer duration than did the children without infection. In comparison with placebo, iron supplementation significantly reduced morbidity due to URTIs in the children with or without infection. In particular, among the children with infection, those who received iron had 29% fewer URTI episodes than did those who received placebo and had infectious episodes that were 40% less severe than those of the children who received placebo. The higher mean (± SD) number of illness-free days in the children who received iron than in those who received placebo (38.3 ± 11.8 compared with 29.0 ± 9.5 d, P < 0.001) suggests that iron supplementation is likely to improve the quality of life of these children and ensure better school attendance.

**TABLE 5** Percentage of children with infection who were treated with oral antibiotics by treatment group

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<th>Antibiotic treatment†</th>
<th>Iron group (n = 127)</th>
<th>Placebo group (n = 52)</th>
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<td>At recruitment (week 0)</td>
<td>56.7 [72]</td>
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*The antibiotic treatment given by the Out Patient Department consisted of 125 mg amoxycillin every 8 h for 3 d. If the child did not show improvement, treatment was given for an additional 3 d.*

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The reduction in morbidity observed in the present study may be due to iron supplementation or the resolution of the infections. The results of statistical analysis indicated that iron supple-mentation had a significant effect apart from the resolution of the infections. Antibiotic treatment cannot solely account for the lower morbidity seen in the iron group than in the placebo group because the number of children who received antibiotics was similar in both the iron and placebo groups at recruitment and during follow-up. Thus, our results suggest that iron supplementation was at least partly responsible for the reduction in morbidity due to URTIs. Note that even the children without infection who received iron had lower morbidity than did their counterparts who received placebo, although the number of URTI episodes and the number of days of illness were low. The number of episodes of gastrointestinal infection was low in the children with or without infection. This may be because the children who were examined in the present study were older than those in previous studies (14, 36) and thus were not as susceptible to frequent diarrhea as were infants and preschool children.

Twenty percent of the children with infection in the present study had a low serum folate concentration; low folate status, however, was not associated with increased morbidity from URTIs because there was no significant difference in morbidity between folate-sufficient (≥3 ng/mL) and folate-deficient children (data not shown). Because a low serum vitamin B-12 concentration was noted in <1% of the children with or without infection, it is unlikely to have been important in the etiology of anemia and infection in this study.

One may speculate that supplementation with iron may be harmful in children who are iron sufficient. However, our analy-sis in the present study indicates that iron supplementation of iron-sufficient children was not associated with adverse effects in terms of infectious morbidity. Previous studies showed impairments in immune function, mainly T cell function, during iron deficiency (6, 7, 38). Iron supplementation may reduce morbidity by improving immune function; additional studies are necessary to establish the mechanisms responsible for the reduction in morbidity observed in the present study.

The strength of the present study is its design, which not only ensured the absence of complications such as malaria, severe protein-energy malnutrition, and other nutrient deficiencies that affect the utilization of iron, but also incorporated the inclusion of an appropriate control group for infection and intervention effects. However, note that in the present study the children with infection were ambulatory outpatients who had recurrent URTIs but were not severely ill. Children who were severely ill with chronic inflammatory conditions and were on long-term treatment with steroids and other drugs were not included in the study. Further studies are needed to determine whether similar benefits would accrue in children who have more chronic or severe inflammatory conditions than those in the present study. A longer duration of intervention and follow-up is also indicated to evaluate the long-term effects of iron supplementation on morbidity.

In conclusion, the results of our study showed that in a community where the prevalence of anemia is high, daily oral iron supple-mentation of 5–10-y-old children for 8 wk improved iron status in children with or without infection. Iron supplementation was also associated with a significant reduction in morbidity due to URTIs and with a greater number of illness-free days. Furthermore, oral iron supplementation did not increase morbidity in iron-sufficient children. Thus, iron supplementation of children with recurrent URTIs should be considered and is likely to be associated with improved iron status, reduced morbidity, and enhanced well-being.

We are grateful to the Ministry of Health, Sri Lanka, for permitting us to carry out this study and to the Director and staff of the Children’s Hospital, Sri Lanka, for their help. We sincerely thank Rajitha Wickremesinghe, Department of Community Medicine, Faculty of Medical Sciences, University of Sri Jayawardanepura, Sri Lanka, for statistical advice. We are grateful to the field investigators for their diligent work and to our subjects and their parents for their participation and cooperation. We thank Sean Lynch for insights and helpful comments.

REFERENCES