In a Randomized Controlled Trial of Iron Fortification, Anthelmintic Treatment and Intermittent Preventive Treatment of Malaria for Anemia Control in Ivorian Children, only Anthelmintic Treatment Shows Modest Benefit1–4

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Abstract
Anemia is common among children in sub-Saharan Africa and its etiology is multifactorial. Likely causes of anemia are low bioavailability of dietary iron, malaria, and helminth infection. In this study, we aimed to assess the effect of iron fortification, intermittent preventive treatment (IPT) of malaria, and anthelmintic treatment on hemoglobin concentration and anemia prevalence among school children. The study was a 6-mo, randomized, double-blind, controlled trial enrolling 591 6- to 24-y-old school children in Côte d’Ivoire using the following: 1) iron-fortified biscuits providing an additional 20 mg iron/d as electrolytic iron 4 times/wk; 2) IPT of malaria with sulfadoxine-pyrimethamine at 0 and 3 mo; and 3) anthelmintic treatment at 0 and 3 mo as the interventions. Prevalence of anemia, iron deficiency, malaria parasitemia, and helminth infection was 70.4, 9.3, 57.7, and 54.8%, respectively. Iron fortification did not improve iron status, IPT of malaria did not affect malaria burden, and neither had an impact on anemia prevalence. Anthelmintics significantly reduced the burden of helminth infections and decreased anemia prevalence (odds ratio: 0.4, 95% CI: 0.3, 0.7). The low prevalence of iron deficiency and an extended dry season that decreased malaria transmission likely reduced the potential impact of iron fortification and IPT. In this setting, anthelmintic treatment was the only intervention that modestly decreased rates of anemia. J. Nutr. doi: 10.3945/jn.109.114256.

Introduction
In the developing world, more than one-half of the school-aged population suffers from anemia; in sub-Saharan Africa alone, there are an estimated 85 million school-aged children afflicted (1). The etiology of anemia is multifactorial, including iron deficiency, malaria, helminth infection, and hemoglobinopathies, as well as other nutritional deficiencies (2). Iron fortification can be an effective strategy to control iron deficiency in developing countries (3), and Côte d’Ivoire has mandated the addition of electrolytic iron to wheat flour (decreed 025, issued on January 18, 2007).

Because inflammation impairs iron absorption and utilization, the efficacy of iron fortification may be blunted in populations in sub-Saharan Africa with high rates of infection.

1Supported by the Medicor Foundation (Vaduz, Liechtenstein), the Swiss Foundation for Research in Nutrition (SFEFS), and the Hochstrasser Foundation (Zurich, Switzerland). In-kind contributions were provided by Midor AG (Meilen, Switzerland; biscuits), Dafra Pharma (Turnhout, Belgium; trial medications), Dr. Lohmann GmbH (Emmerthal, Germany; electrolytic iron), and Nestlé (Abidjan, Côte d’Ivoire; climate chambers for biscuit storage). The sponsors of the study played no role in the design or implementation of the trial reported, analysis of the data, or the preparation, submission, and revision of the manuscript.


3This trial was registered at controlled-trials.com (ISRCTN21782274).

4Supplemental Figure 1 is available with the online posting of this paper at jn.nutrition.org.

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Thus, it may be necessary to control parasite infections to reduce inflammation and ensure iron bioavailability from fortified foods. Large-scale administration of anthelmintic drugs in areas highly endemic for helmintic infections (e.g. lymphatic filariasis, onchocerciasis, schistosomiasis, and soil-transmitted helmintiasis) is increasingly promoted by the WHO (5). Intermittent preventive treatment (IPT) of malaria with sulfadoxine-pyrimethamine (SP)\(^\text{12}\) significantly reduced malaria parasitemia and morbidity in pregnant women and infants (6,7). The latter strategy may also improve health and cognitive performance in school-aged children (8).

Therefore, our aim in this study was to assess the relative efficacy of iron fortification, IPT of malaria using SP, and anthelmintic treatment with albendazole plus praziquantel, administered alone or in combination, to improve hemoglobin (Hb) levels and reduce anemia in school-aged children in rural Africa.

**Participants and Methods**

**Study area and participants.** The study population consisted of 6- to 14-y-old school children in 5 villages located 15–20 km south of Toumodi in central Côte d’Ivoire. The area is in the transition zone from rainforest to savannah with an annual mean temperature of 27°C. The wet season usually lasts from March to November, with a dry spell in the months of July and August, and the heaviest rains occurring between March and May.

**Study design, sample size, and ethics.** This was a double-blind, randomized, placebo-controlled trial, using a 2 × 2 × 2 factorial design lasting 6 mo. Field work was carried out between November 2006 and July 2007. Assuming a mean Hb of 117 ± 12 g/L (9) and that an increase of 8 g/L in Hb would be clinically relevant, and allowing for a drop-out rate of 20%, we enrolled 80 children per group to achieve a power of 90% at a 5% level of significance.

Approval was given by the ethical review boards of the ETH Zurich, Switzerland (2006–23), the University of Basel (EKBB), Switzerland (224/06), and the Ministry of Health in Côte d’Ivoire (5782/MSHP/CAB/CNESVS/06). Village authorities, parents, and legal guardians of eligible children were informed about the purpose, procedures, and potential risk and benefits of the study. Written informed consent was obtained from parents or legal guardians of the participating children.

**Randomization and interventions.** Inclusion criteria for girls were nonpregnant (self-reported) and for boys and girls were: no major chronic illnesses; anticipated local residence for the study duration; known or reported hypersensitivity to SP, albendazole, or praziquantel; no acute febrile illness; no known or reported infections with helminths, Plasmodium, or Treponema pallidum; no known or reported HIV infection; no known or reported immunocompromising conditions; no known or reported malignancies; no known or reported local residence for the study duration; no known or reported severe local environment; and no anthelmintic treatment within the previous 4 wk. Children were individually assigned to 1 of 8 groups of all possible combinations of the 3 interventions: 1) iron fortification; 2) IPT of malaria; and 3) anthelmintic treatment or the respective placebo (Table 1).

The iron fortification groups received 2 fortified “petit beurre” type biscuits [Midor AG; electrolytic Fe (A-131; Dr. Lohmann), 20 mg Fe/dl] 4 times/wk. Placebo recipients consumed the same biscuits but unfortified. Iron content of the biscuits was measured using atomic absorption spectrometry (Spectra AA-50, Varian). The 2 types of biscuits were compared in a triangle test (10) and were indistinguishable by local adults (n = 24; data not shown). Children assigned to IPT of malaria received SP (500 mg sulfadoxine plus 25 mg pyrimethamine) given after the baseline screening and 3 mo later. Placebo recipients were administered a matching placebo. The anthelmintics group was given albendazole (single 400-mg oral dose) plus praziquantel (single 40-mg/kg oral dose) after the baseline screening and 3 mo later. Placebo recipients were given a matching placebo. Trial medications and placebo were provided by Dafra Pharma. Groups were coded by 8 letters, each coding for 1 of the 8 treatment combinations. Children were individually randomized to 1 of the randomization letters in blocks of 8. The codes were held by a member of an independent data safety and monitoring board until data analysis was completed.

**Laboratory methods.** Biomedical variables were assessed at baseline and endpoint. Venous blood (6 mL) was drawn into EDTA-coated Vacutainer tubes (Becton Dickinson). Blood was transported on ice to the nearby laboratory. Hb was measured using an AcT8 Counter (Beckman Coulter) on the day of blood sampling; anemia was defined according to WHO criteria (1). Subsequently, Giemsa-stained thick and thin blood films were examined microscopically and Plasmodium parasites were counted against 200–500 leukocytes and converted to the number of parasites/µL of blood, assuming a leukocyte count of 8000/µL (11). Parasitemia levels were defined as the following ranges: 0; 1–999; 1000–1999; 2000–4999; and ≥5000 parasites/µL of blood (12). Hb typing was done by electrophoresis in a subsample of 110 randomly selected children (13).

Plasma was aliquoted, transported frozen, and stored at ~25°C for analysis of plasma ferritin (PF), soluble transferrin receptor (TIR), C-reactive protein (CRP), and α-1-acid-glycoprotein (AGP). Zinc protoporphyrin (ZPP) was measured on washed RBC using a hemato-fluorometer (Aviv Biomedical) within 7 d after sampling. PF and CRP were measured using an automated chemiluminescent immunoassay system (IMMULITE, Diagnostic Products). TIR was measured using an automated immunonephelometric assay (Cobas Integra 800, Roche Diagnostics). AGP was measured by immunoturbidimetry (Cobas Mira, Roche Diagnostics). Normal reference values are: ZPP, <40 µmol/mol heme; TR, >8.5 µg/L; and PF, >30 µg/L (to convert from µg/L to pmol/L, multiply by 2.247) (14). Anemia was defined as Hb <8.0 g/dL and <115 g/L for children <12 y and ≥8.0 g/dL and ≤120 g/L for children >12 y (1). Iron deficiency was defined as [PF <30 µg/L or (TIR >8.5 µg/L and ZPP >40 µmol/mol heme)] (15). Inflammation was defined as AGP >1.2 g/L or CRP >10 mg/L (16).

Two Kato-Katz thick smears (41.7 mg each) were prepared from each stool sample and hookworm eggs were counted under a microscope (17). The slides were reexamined and the number of eggs of Ascaris lumbricoides, Schistosoma mansoni, and Trichuris trichiura were counted. For the conversion to eggs/g feces (EPG), a multiplication factor of 24 was employed. Criteria for light infections with A. lumbricoides (<5000 EPG), T. trichiura (<1000 EPG), hookworm (<2000 EPG), and S. mansoni (<100 EPG) were according to WHO guidelines (18).

**Statistical analysis.** Data analysis was conducted in STATA version 9 and Excel (2003 issue). All analyses were conducted using a modified

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12 Abbreviations used: AGP, α-1-acid-glycoprotein; CRP, C-reactive protein; EPG, eggs/g feces; FMH, iron fortification-malaria intervention-helmintihm intervention group; FMP, iron fortification-malaria intervention-placebo group; FPP, iron fortification-placebo-malaria intervention group; FPH, iron fortification-placebo-helmintihm intervention group; Hb, hemoglobin; IPT, intermittent preventive treatment of malaria; OR, odds ratio; PF, plasma ferritin; PMH, placebo-malaria intervention-helmintihm intervention group; PMP, placebo-malaria intervention-placebo group; PPH, placebo-placebo-malaria intervention group; PPP, placebo-placebo-placebo group; SP, sulfadoxine-pyrimethamine; TIR, soluble transferrin receptor; ZPP, zinc protoporphyrin.
intention-to-treat approach, imputing “median values of own group” for missing endpoint values ($imputed = 37; 6.3\%$) and excluding participants from analysis who did not have at least the baseline assessment. Missing birth dates were imputed by calculating the mean age per school grade ($imputed = 77; 13.0\%$). Between-group comparisons for baseline values, as well as within-group across time point comparisons, were performed using 1-way ANOVA for normally distributed data, and Kruskall-Wallis test for skewed continuous data; chi-square test or Fisher’s exact test, as appropriate, for binary data; and negative binomial regression for overdispersed egg or parasite counts. Multiple regression models were applied to assess the effects of iron, IPT, and anthropometric treatment and their interactions at endpoint on differences in Hb as primary outcome, log-transformed PF, TTR, and ZPP (linear regressions), hookworm intensities, and Plasmodium parasitemia (negative binomial regressions). For example, we estimated the effect of iron fortification by comparing the groups that received iron (i.e. iron fortification-placebo-placebo (FPP), iron fortification-malaria intervention-helminth intervention (FPH), iron fortification-malaria intervention-placebo (FMP), and iron fortification-malaria intervention-helminth intervention (FMH)) with the groups that did not receive iron (i.e. placebo-placebo-placebo (PPP), placebo-placebo-helminth intervention (PPH), placebo-malaria intervention-placebo (PMP), and placebo-malaria intervention-helminth intervention (PMH)), after adjusting for the effect of potential confounders, such as respective baseline parameter, inflammation, sex, age, and biscuit intake compliance. Logistic models were fitted to perform the above regression analyses on the binary variables of anemia, iron deficiency, malaria, hookworm, and inflammation prevalence [odds ratio (OR), including 95% CI]. Significance was assessed at 5% significance level using the F-test (linear regressions) and likelihood ratio test (logistic and negative binomial regressions). Values in the text are means ± SD or geometric mean (95% CI) unless noted otherwise.

**Results**

A total of 591 school children were enrolled and 554 completed the study (Supplemental Fig. 1). The main reason for drop-out was out-migration due to a persisting teachers’ strike. In-trial reasons for exclusion from the study but not from analysis were: PPP: splenomegaly ($n = 1$); FPP: endocrinological disease ($n = 1$) and refusal to consume biscuits ($n = 1$); PMP: adenopathy ($n = 1$); FPH: adenopathy ($n = 1$) and pregnancy ($n = 1$); and FMH: massive hematuria ($n = 1$) and pneumonia/respiratory disease ($n = 1$).

Table 2 shows the baseline characteristics of the study population; groups were comparable for all the assessed parameters. The age of the children was $9.8 ± 2.5 \text{ y}$; $42.8\%$ were females. Their Hb concentration was $111.4 ± 9.9 \text{ g/L}$ and anemia prevalence was $70.4\%$, but only $9.3\%$ were iron deficient. Of the children in the trial, $54.8\%$ were infected with any kind of helminth species, with hookworm being the predominant species; the number of children infected with hookworm, *T. trichiura*, and *A. lumbricoides* was 311, 17, and 8, respectively. The geometric mean infection intensity of hookworm was 107.8 EPG (95% CI: 92.4, 125.7). Overall, infection intensity was low; only 5 children (1.6\%) had a hookworm infection intensity $>2000$ EPG and no child exceeded the WHO cut-off values for light infection intensities for *A. lumbricoides* and *T. trichiura*. More than one-half of the children (57.7\%, $n = 341$) were infected with *Plasmodium* spp. The geometric mean of *Plasmodium*-positive children was 373.6 parasites/µL of blood (95% CI: 321.3, 434.5). Parasitemia was generally low; among the infected children, 70.1\% had $<1000$ parasites/µL of blood, 17.6\% between 1000 and 1999, 8.2\% between 2000 and 4999, and 3.2\% had a high parasitemia ($\geq 5000$).
### TABLE 3  Endpoint characteristics and differences from baseline of enrolled Ivorian children by treatment group

<table>
<thead>
<tr>
<th></th>
<th>PPP</th>
<th>FPP</th>
<th>PPH</th>
<th>PMP</th>
<th>FPH</th>
<th>PMH</th>
<th>FMP</th>
<th>FMH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (g/L)</td>
<td>106.7 ± 9.4</td>
<td>107.2 ± 9.2</td>
<td>109.6 ± 9.2</td>
<td>107.9 ± 10.4</td>
<td>109.3 ± 10.7</td>
<td>109.7 ± 9.2</td>
<td>107.8 ± 8.7</td>
<td>109.3 ± 9.0</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>−3.9 (−6.1, −1.8)</td>
<td>−3.9 (−6.5, −1.4)</td>
<td>−1.2 (−3.2, 0.7)</td>
<td>−4.5 (−6.9, −2.1)</td>
<td>−1.9 (−3.2, 0.8)</td>
<td>−1.4 (−3.6, 0.7)</td>
<td>−5.4 (−7.3, −3.5)</td>
<td>−1.2 (−3.4, 1.0)</td>
</tr>
<tr>
<td>Pt (µg/L)</td>
<td>63.9 (41.1, 82.9)</td>
<td>60.6 (40.7, 72.7)</td>
<td>63.3 (48.1, 80.0)</td>
<td>58.8 (35.9, 81.9)</td>
<td>66.5 (47.6, 88.3)</td>
<td>69.3 (45.1, 87.6)</td>
<td>54.0 (36.4, 81.2)</td>
<td>68.3 (45.6, 88.8)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>−15.5 (−33.3, 4.2)</td>
<td>−5.6 (−21.2, 10.6)</td>
<td>−15.5 (−34.2, 0.0)</td>
<td>−5.1 (−16.0, 6.5)</td>
<td>3.7 (−8.4, 13.1)</td>
<td>3.5 (−23.9, 17.6)</td>
<td>−2.4 (−25.0, 12.9)</td>
<td>−0.6 (−11.1, 9.9)</td>
</tr>
<tr>
<td>TR (mg/L)</td>
<td>5.9 (4.5, 7.2)</td>
<td>5.9 (5.0, 7.5)</td>
<td>5.9 (5.2, 7.1)</td>
<td>5.8 (4.9, 7.3)</td>
<td>6.0 (5.3, 7.7)</td>
<td>6.0 (4.6, 7.2)</td>
<td>5.9 (4.5, 7.3)</td>
<td>6.4 (5.2, 7.3)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>2.0 (1.0, 3.1)</td>
<td>2.1 (1.2, 3.3)</td>
<td>2.1 (1.3, 2.9)</td>
<td>2.2 (1.1, 3.2)</td>
<td>2.1 (1.0, 3.2)</td>
<td>2.0 (0.9, 3.2)</td>
<td>1.8 (0.9, 2.9)</td>
<td>2.2 (1.3, 3.0)</td>
</tr>
<tr>
<td>ZPP (µmol/mol)</td>
<td>5.3 (43.0, 73.5)</td>
<td>5.3 (39.0, 69.5)</td>
<td>5.2 (37.3, 69.5)</td>
<td>5.0 (37.8, 58.5)</td>
<td>5.3 (40.0, 66.5)</td>
<td>4.9 (41.0, 56.5)</td>
<td>5.1 (42.0, 63.0)</td>
<td>4.7 (40.0, 54.0)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>0.0 (−9.0, 10.0)</td>
<td>−2.5 (−10.5, 12.0)</td>
<td>−0.5 (−7.3, 8.3)</td>
<td>−1.0 (−11.0, 8.5)</td>
<td>−1.0 (−11.0, 8.5)</td>
<td>−5.5 (−13.0, 2.0)</td>
<td>−4.3 (−10.0, 4.5)</td>
<td>−5.0 (−17.8, 4.5)</td>
</tr>
<tr>
<td>Parasitism (%)</td>
<td>564 (359, 887)</td>
<td>614 (349, 1079)</td>
<td>605 (316, 1161)</td>
<td>822 (478, 1416)</td>
<td>767 (454, 1298)</td>
<td>688 (386, 1227)</td>
<td>810 (417, 1572)</td>
<td>651 (357, 1187)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>0 (−224, 112)</td>
<td>0 (−448, 128)</td>
<td>−40 (−296, 0)</td>
<td>0 (−144, 560)</td>
<td>0 (−176, 0)</td>
<td>0 (−420, 0)</td>
<td>0 (−128, 176)</td>
<td>0 (−400, 0)</td>
</tr>
<tr>
<td>Hookworm infection intensity (EPG)</td>
<td>355 (233, 540)</td>
<td>367 (250, 538)</td>
<td>79 (41, 154)</td>
<td>408 (294, 566)</td>
<td>130 (77, 219)</td>
<td>130 (75, 224)</td>
<td>375 (264, 531)</td>
<td>98 (49, 198)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>72 (0, 708)</td>
<td>168 (0, 540)</td>
<td>0 (−48, 0)</td>
<td>228 (0, 612)</td>
<td>0 (−96, 0)</td>
<td>0 (−156, 0)</td>
<td>180 (0, 576)</td>
<td>0 (−60, 0)</td>
</tr>
<tr>
<td>Anemia (%)</td>
<td>65 (87.8)</td>
<td>65 (89.0)</td>
<td>48 (70.6)</td>
<td>64 (84.2)</td>
<td>59 (78.7)</td>
<td>58 (76.3)</td>
<td>67 (85.9)</td>
<td>54 (76.1)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>148 (31.2, 25.6)</td>
<td>151 (4.7, 25.9)</td>
<td>4.4 (−9.6, 18.4)</td>
<td>17.1 (59.2, 83.3)</td>
<td>0.8 (−42, 20.2)</td>
<td>6.5 (−75, 20.7)</td>
<td>20.5 (9.5, 31.4)</td>
<td>−1.4 (15.5, 12.6)</td>
</tr>
<tr>
<td>Iron deficiency (%)</td>
<td>13 (17.6)</td>
<td>15 (20.8)</td>
<td>9 (13.4)</td>
<td>15 (19.7)</td>
<td>16 (21.3)</td>
<td>10 (13.3)</td>
<td>17 (21.8)</td>
<td>11 (15.5)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>42 (−5.4, 13.9)</td>
<td>11.1 (0.6, 21.6)</td>
<td>10.6 (−0.4, 21.6)</td>
<td>7.8 (−0.2, 18.0)</td>
<td>162 (56.6, 28.6)</td>
<td>4.1 (−60, 14.1)</td>
<td>11.8 (0, 223)</td>
<td>5.7 (−35, 14.9)</td>
</tr>
<tr>
<td>Inflammation (%)</td>
<td>16 (22.2)</td>
<td>16 (23.2)</td>
<td>15 (22.4)</td>
<td>13 (17.3)</td>
<td>18 (24.0)</td>
<td>15 (20.3)</td>
<td>16 (21.1)</td>
<td>14 (20.0)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>−7.1 (−21.3, 7.0)</td>
<td>−6.1 (−22.1, 9.9)</td>
<td>−9.0 (−25.2, 7.3)</td>
<td>−17.6 (−31.5, −36)</td>
<td>−1.4 (−15.4, 12.7)</td>
<td>−2.8 (−17.7, 12.1)</td>
<td>−1.3 (−14.1, 11.4)</td>
<td>−1.3 (−28.5, 2.8)</td>
</tr>
<tr>
<td>Malaria (%)</td>
<td>29 (39.2)</td>
<td>27 (37.0)</td>
<td>21 (30.9)</td>
<td>31 (40.8)</td>
<td>21 (28.0)</td>
<td>23 (30.3)</td>
<td>28 (35.9)</td>
<td>23 (32.4)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>−23.0 (−38.1, −7.8)</td>
<td>−16.4 (−34.3, 15)</td>
<td>−27.9 (−44.0, −11.9)</td>
<td>−17.1 (−32.7, −15)</td>
<td>−30.7 (−45.3, −16.0)</td>
<td>−28.9 (−42.8, −5.1)</td>
<td>−16.7 (−32.8, −0.6)</td>
<td>−26.8 (−43.3, −10.3)</td>
</tr>
<tr>
<td>Hookworms (%)</td>
<td>52 (70.3)</td>
<td>57 (78.1)</td>
<td>18 (26.5)</td>
<td>58 (73.3)</td>
<td>16 (21.3)</td>
<td>7 (9.2)</td>
<td>60 (76.9)</td>
<td>14 (19.7)</td>
</tr>
<tr>
<td>Difference (%)</td>
<td>19.2 (66.3, 31.7)</td>
<td>23.3 (10.8, 35.7)</td>
<td>−29.4 (−45.0, −13.8)</td>
<td>18.7 (42.3, 6.9)</td>
<td>−32.0 (−46.7, −17.3)</td>
<td>−40.8 (−55.3, −26.2)</td>
<td>19.2 (7.8, 30.6)</td>
<td>−22.5 (−36.4, −8.7)</td>
</tr>
</tbody>
</table>

1. Data are mean ± SD or n (%) unless indicated otherwise.
2. Absolute endpoint value.
3. Mean difference endpoint vs. baseline survey (95% CI), paired t test.
4. Median (IQT).
5. Median difference endpoint vs. baseline survey (IQT), Wilcoxon's Signed Rank test.
6. Geometric mean (95% CI) of positive individuals.
7. Negative binomial regression.
8. Percent difference endpoint vs. baseline survey (95% CI), McNemar’s χ² test; *significant change from baseline within the own group, P < 0.05.
At baseline, 28% of the children showed biochemical signs of inflammation and the OR for anemia or for inflammation was 2.3 (95% CI: 1.5, 3.5). Of the children screened for hemoglobinopathies, 86.4% had normal genotype (HbAA), followed by HbAS (6.3%), HbAC (3.6%), HbAF (1.8%), HbCC (0.9%), and HbSC (0.9%).

During the trial, the number of biscuits consumed was 164.2 ± 21.7 of 200 biscuits offered (overall biscuit intake compliance rate: 82.1%) with no difference among groups (P = 0.53). The iron content of the biscuits was 9.6 ± 0.3 mg. Thus, total additional iron consumed by the children receiving the fortified biscuits was 1576 ± 216 mg of electrolytic iron. This corresponds to a daily intake of 9.0 ± 1.2 mg, more than 3 times the estimated daily iron dose provided by the Ivorian flour fortification program for this age group. Compliance to IPT of malaria and anthelmintics was 93.7%; 3 children were administered the wrong drugs. Post-trial, 2 of these 3 children could be reassigned to a new, matching treatment group.

During the study, in the PPP group, there was a significant decrease in Hb of 3.9 g/L (95% CI: −6.1, −1.8), anemia prevalence increased, TIR increased, and PF concentrations decreased (Table 3). Both malaria prevalence and parasitemia decreased in all groups, whereas hookworm prevalence and egg burden increased in those participants not receiving anthelmintic treatment, and TIR increased in all groups.

Analysis for the main effects revealed that anthelmintic treatment was highly efficacious in reducing hookworm infection (Table 4). In contrast, IPT of malaria did not significantly reduce malaria parasitemia and iron fortification did not affect iron deficiency or any of the iron status indicators. Consequently, only anthelmintic treatment resulted in a modestly increased Hb concentration by 2.4 g/L (95% CI: 1.2, 3.7) and a lower risk for anemia, as indicated by an OR of 0.4 (95% CI: 0.3, 0.7). Moreover, in the groups receiving anthelmintics, all iron status indicators improved (P ≤ 0.03). In the groups receiving IPT of malaria, ZPP improved (P < 0.01), whereas hookworm infection intensity was reduced (P = 0.03). There were no significant interactions among any of the 3 interventions (Table 4).

**Discussion**

We investigated the effect of 3 interventions or combinations thereof, iron fortification, IPT of malaria, and anthelmintic treatment, to improve Hb levels and reduce anemia among school-aged children in Africa. Anthelmintic treatment was the only intervention that had a significant, but only modest, beneficial effect on both Hb concentration and anemia rates.

Hb and iron status of the PPP group, assessed by PF, TIR, and ZPP, deteriorated during the 6-mo study period. At the same time, malaria parasitemia decreased. A possible explanation of these findings is an unusual delay (by nearly 3 mo) in the onset of the rainy season during the study. The prolonged dry season reduced dietary variety and may have decreased dietary iron bioavailability due to reduced availability of meat, fish, and poultry, and increased reliance on cereals high in phytic acid. Due to the prolonged dry season, most of the standing water disappeared, which likely reduced malaria transmission (19).

Despite the high prevalence of mostly mild anemia, only 9% of the children were iron deficient. Previous studies in children living in rural parts of Côte d’Ivoire have reported anemia prevalences between 36 and 50%, lower than in our study, but reported iron deficiency prevalences of 23–35%, higher than in our study (12,20). These differences may be due to variations in diet, rates of infectious diseases, prevalence of hemoglobinopathies such as α-thalassemia, as well as the recognized difficulty of accurately defining iron deficiency in sub-Saharan Africa (14).

The low prevalence of iron deficiency may have reduced the effect of the iron fortification, because fractional absorption of dietary iron is inversely related to body iron stores. However, a mean of >1.5 g of electrolytic iron was consumed during the study period in the groups receiving the iron-fortified biscuits. Even if only a small fraction of this iron dose had been absorbed, it should have been visible as an increase in storage iron (PF); iron stores can be increased by iron fortification in Ivorian children who have an adequate iron status (20). The reason for the negligible iron absorption in this study is unclear. The biscuits were made from low-extraction flour and were given to children at the morning breaks at schools, and there was no

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### Table 4: Results of the main effects and interactions of the trial according to the 3 interventions (iron fortification, anthelmintic treatment, and IPT of malaria)

<table>
<thead>
<tr>
<th></th>
<th>Iron fortification</th>
<th>Anthelmintics</th>
<th>IPT</th>
<th>Main effects</th>
<th>Interactions</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Estimate</td>
<td>P</td>
<td>Estimate</td>
<td>P</td>
<td>Estimate</td>
</tr>
<tr>
<td>Hb, g/L</td>
<td>1.2</td>
<td></td>
<td></td>
<td>&lt;0.01</td>
<td>0.68</td>
</tr>
<tr>
<td>Log (PF)</td>
<td>0.00±0.04</td>
<td>0.12</td>
<td>&lt;0.01</td>
<td>0.06</td>
<td>0.18</td>
</tr>
<tr>
<td>Log (TIR)</td>
<td>0.00±0.00</td>
<td>−0.14</td>
<td>0.03</td>
<td>&lt;0.07</td>
<td>0.30</td>
</tr>
<tr>
<td>Log (ZPP)</td>
<td>0.00±0.00</td>
<td>−0.07</td>
<td>&lt;0.01</td>
<td>0.07</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Malaria parasitemia, parasites/μL blood</td>
<td>1.59±0.22</td>
<td>0.57</td>
<td>0.00</td>
<td>1.46</td>
<td>0.34</td>
</tr>
<tr>
<td>Hookworm infection intensity, EPG</td>
<td>0.59±0.92</td>
<td>0.04</td>
<td>&lt;0.01</td>
<td>0.08</td>
<td>0.03</td>
</tr>
<tr>
<td>Anemia, %</td>
<td>1.1±1.06</td>
<td>0.41</td>
<td>&lt;0.01</td>
<td>0.99</td>
<td>0.98</td>
</tr>
<tr>
<td>Iron deficiency, %</td>
<td>1.42±0.14</td>
<td>0.81</td>
<td>0.37</td>
<td>0.84</td>
<td>0.46</td>
</tr>
<tr>
<td>Inflammation, %</td>
<td>1.12±0.60</td>
<td>1.10</td>
<td>0.72</td>
<td>0.86</td>
<td>0.47</td>
</tr>
<tr>
<td>Malaria, %</td>
<td>0.94±0.75</td>
<td>0.70</td>
<td>0.05</td>
<td>1.15</td>
<td>0.45</td>
</tr>
<tr>
<td>Hookworms, %</td>
<td>1.25±0.29</td>
<td>0.05</td>
<td>&lt;0.01</td>
<td>0.83</td>
<td>0.38</td>
</tr>
</tbody>
</table>

1. Mean estimate.
2. P-value is based on F-test statistic.
3. Density rate ratio.
4. P-value is based on likelihood ratio test statistic.
5. OR.
other food intake for the next 2 h. Thus, dietary inhibitors of iron absorption, such as phytic acid and/or polyphenols, are unlikely to have played a role. Three of 4 controlled trials using electrolytic iron at varying doses in infants or school-aged children in sub-Saharan Africa have not been efficacious (21–24). In contrast, 10 mg/d electrolytic iron fed to Thai women with low iron stores for 8 mo was efficacious (25). These differences are likely due to a higher frequency of infections in African populations that reduce iron absorption and/or utilization. In an efficacy study of iron-fortified maize in Kenyan children, sodium iron EDTA reduced rates of iron deficiency anemia, whereas electrolytic iron did not (24). Recent guidelines recommend sodium iron EDTA as the fortificant of choice in populations with high infection rates and a relatively low consumption of wheat flour (26).

In contrast to clear guidelines for IPT of malaria in pregnant women and infants in stable transmission areas (27,28), there is currently no consensus on an IPT regimen for school-aged children. The IPT regimen used in this study was likely ineffective for several reasons: 1) 2 doses of SP appropriate for a child of 20 kg were administered, but this dose may have been too low in our older and therefore heavier children; 2) resistance to SP has recently been reported for neighboring countries of Côte d’Ivoire (29); and 3) the atypically long dry season during the study may have reduced malaria transmission. However, the overall decrease in malaria parasitemia during this dry period had no measurable beneficial impact in our study population neither on Hb status nor on anemia.

The 2 single-dose treatment courses with albendazole, administered at a 3-mo interval, were highly efficacious in reducing hookworm burden and improving anemia rates in this setting where most children carry light-intensity hookworm infections. The benefit of anthelmintics on anemia rates is thought to be due to reduced iron losses from intestinal bleeding as well as improved iron utilization due to reduced inflammation (30,31). Although a reduction of inflammation (as measured using CRP and AGP) could not be shown, our findings confirm the iron status-mediated pathway leading to increased Hb levels; all 3 iron indicators were significantly improved in the groups receiving anthelmintics.

Our findings emphasize that strategies to control anemia in children in regions of sub-Saharan Africa should be based on careful delineation of the local causes of anemia. Anthelmintic drugs are a simple-to-implement public health strategy that may benefit populations where hookworm infections are pervasive, as in this study. In contrast, IPT of malaria with SP in this setting was not beneficial, but more effective drugs for IPT have since become available. Due to the low prevalence of iron deficiency and the high prevalence of infections in rural Côte d’Ivoire, iron fortification, particularly with a poorly soluble compound like electrolytic iron, is unlikely to be effective in reducing anemia rates in children. Regular administration of anthelmintic drugs (i.e. albendazole plus praziquantel) to school-aged children, however, is likely to improve anemia, on top of reducing morbidity due to soil-transmitted helminthiasis and schistosomiasis.

Acknowledgments
We thank Adjoba M. Brou-Tanoh, Julien Donnio, and Thomas A. Smith for serving on the data safety and monitoring board; Hala Ghatts for assistance in field work and supervision; Mahamadou Bakayoko, Kouadio J. Brou, Sostène Brou, Kouassi N. Lingué, Laurent K. Lohourignon, Moussan N’Cho, Diabaté Saulo, Kigbafori D. Silué, Mahamadou Traoré, and Evelyne N. Yapi for technical assistance in parasitology; Ursula B. Bouah, Marcelle F. Dibi Ahou, T’chan Djé Bi, Laure-Edito O. Klognigué, Eric S. Kubetzerić, Tanoh N’Diama, Boroba F. N’Douba, Guillaume Y. N’Dri, Kouakou Ossé, and Nalégou B. Yéo for field work. F.R., M.B.Z., P.V., J.U., and R.F.H. designed research; F.R., R.J.A., A.B.T., E.K.N., M.C., M.D.T., H.A., D.E.S., and C.N. conducted research; F.R., M.B.Z., P.V., J.U., and R.F.H. analyzed and interpreted data; F.R., M.B.Z., J.U., and R.F.H. wrote the paper; and F.R. had primary responsibility for final content. All authors read and approved the final submitted and revised manuscript.

Literature Cited


