

Treatment of anemia with microencapsulated ferrous fumarate plus ascorbic acid supplied as sprinkles to complementary (weaning) foods¹⁻³

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ABSTRACT

Background: Standard therapy for anemia in infants is ferrous sulfate drops administered 3 times/d. Adherence to treatment, however, is often poor. One likely reason for poor adherence is the unpleasant side effects associated with drops.

Objective: The objective was to evaluate the use of a new form of iron and a delivery system to treat anemia in infants that is likely to produce better adherence to treatment.

Design: Using a prospective, randomized, controlled design, we studied 557 anemic children aged 6–18 mo (hemoglobin: 70–99 g/L) in rural Ghana. One group received a daily sachet of microencapsulated ferrous fumarate (80 mg elemental Fe) in powder form plus ascorbic acid to be sprinkled onto any complementary food eaten (sprinkles group); a control group received ferrous sulfate drops 3 times/d for 2 mo (total dose: 40 mg elemental Fe). Hemoglobin and serum ferritin concentrations were measured at baseline and at the end of treatment.

Results: Successful treatment of anemia (hemoglobin > 100 g/L) occurred in 58% of the sprinkles group and in 56% of the drops group, with minimal side effects in both groups. Geometric mean ferritin concentrations increased significantly in each group from baseline to the end of treatment ($P < 0.001$).

Conclusion: Use of ferrous sulfate drops or a single daily dose of microencapsulated ferrous fumarate sprinkles plus ascorbic acid resulted in a similar rate of successful treatment of anemia without side effects. To our knowledge, this is the first demonstration of the use of microencapsulated iron sprinkles to treat anemia. Improved ease of use may favor the use of sprinkles to deliver iron. *Am J Clin Nutr* 2001;74:791–5.

KEY WORDS Iron, infants, children, anemia, microencapsulated iron, ferrous fumarate, ferrous sulfate drops, Ghana

INTRODUCTION

Although major advances have been made in the treatment and prevention of vitamin A and iodine deficiency over the past 10 y, there has been a marked lack of success in the prevention of iron deficiency and iron deficiency anemia. Current estimates suggest that as many as one-third to one-half of all individuals in the developing world are iron deficient or anemic (1). In 1996, a group of UNICEF consultants reviewed possible interventions to treat and prevent anemia. It was quite clear then (and

now) that available interventions (syrup and drops for infants and children, and capsules for women) are not effective (2). For many reasons, adherence to treatment is poor despite multiple efforts to influence and improve it (3). At that meeting, a new method to provide micronutrients (including iron) to populations at risk was suggested. The intervention was based on 2 observations from the West, where micronutrient deficiencies are rare (4). The observations are that 1) commercial food fortification works well to prevent deficiencies, and 2) the fortifi-cants must not appreciably change the color, texture, or taste of the food to which they are added.

Our suggested approach was to encapsulate micronutrients so that they could be added directly to food at the table. Pharmaceutical encapsulation has been used for years to mask the strong taste of certain drugs and even vitamins (5). The encapsulate is a thin coating of a soya-based hydrogenated lipid. Encapsulation prevents the micronutrients from oxidizing the food, which means that there is no change in the color or taste of the food. To administer the correct amount of iron, the encapsulated micronutrients were packaged in single-dose sachets, as is the case with oral rehydration sachets. To use the sachets, one is instructed to sprinkle the entire contents onto whatever food is being served in the household. The research described in this manuscript is the initial field testing of the sprinkles.

In this prospective, randomized, controlled trial, we tested the hypothesis that the response to treatment of anemia would be better with 2 mo of treatment with microencapsulated ferrous fumarate sachets daily than with ferrous sulfate drops provided 3 times/d.

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SUBJECTS AND METHODS

Study area, subjects, and recruitment

The study took place between May and August 1999 in the field study area for the Kintampo Health Research Centre, located in the Kintampo district of Ghana. This is a malaria-endemic area where the principal complementary food is a maize-based porridge. The prevalence of anemia in young children is estimated to be $\approx 70\%$, a significant proportion of which is due to iron deficiency (6).

Eligible infants were identified from an existing surveillance database of births in the district. To be included in the study, infants had to be 6–18 mo of age at the time of recruitment, be ingesting a weaning food in addition to breast milk, and have a hemoglobin concentration between 70 and 99 g/L, measured during a baseline assessment. Children who were severely anemic (hemoglobin < 70 g/L) were excluded from the trial and treated.

Ethics approval for this study was obtained from the Research Ethics Committees at The Hospital for Sick Children (Toronto), the London School of Hygiene and Tropical Medicine (London), and Ghana's Ministry of Health through the Health Research Unit (Kintampo, Ghana).

Oral consent to conduct the study in the Kintampo district was obtained from the District Assembly of Elected Representatives. Consent to conduct the study in each village was obtained from village elders, and individual consent to participate in the study was obtained from the mothers of the infants included in the study.

Study design

Because it would be unethical to provide a placebo to a child with anemia, we did not include a placebo control. After the baseline assessment, children were randomly assigned to 1 of 2 treatment groups. Randomization was done with sealed opaque envelopes containing group designations, which were generated randomly by computer with Microsoft Access 97 (Microsoft Corporation, Seattle). It was not feasible to blind the field staff or the mothers to the group to which the children were assigned. However, the persons responsible for the laboratory and data analyses were blinded to the group designations.

The drops group received ferrous sulfate drops (5 mg elemental Fe \cdot kg⁻¹ \cdot d⁻¹, rounded to a total of 40 mg elemental Fe) provided in 3 equal doses/d (7). The sprinkles group received microencapsulated ferrous fumarate (80 mg elemental Fe) packaged in a sachet with ascorbic acid (50 mg), which was added to the child's meal (after it was cooked) once daily. The sprinkles were mixed and packaged in Toronto and shipped by air to Ghana. A single bottle of ferrous sulfate was dispensed to mothers in the drops group once every 4 wk. Mothers in the sprinkles group were provided with an adequate number of sachets for 2 wk.

The dosage of iron in the sprinkles sachet was double that in the ferrous sulfate drops. On the basis of algorithms, which account for major enhancers and inhibitors of iron absorption, we estimated that the absorption of encapsulated iron (in the sachets) would be significantly lower than that of the nonencapsulated iron (8–10). However, to our knowledge, no previous studies in humans have examined the absorption of hydrogenated soy lipid-coated ferrous fumarate sprinkles. A recent study by Davidsson et al (11) reported that the mean absorption in infants of iron from standard ferrous fumarate at fortification levels in infant cereal was 4.1% (range: 1.7–14.7%). These data cannot be directly extrapolated to iron absorption from encapsulated ferrous fumarate for many reasons.

The encapsulation matrix (hydrogenated soy lipid) may have some effect on the dissolution properties of ferrous fumarate; Davidsson et al's test meal (white wheat- and soy-based flours) was relatively low in phytic acid, a major inhibitor of iron absorption, whereas in Ghana, the phytate content of cereals is very high (12). In the current study, infants were anemic, suggesting higher absorption potential. However, we used Davidsson et al's data in our algorithms, which estimated absorption of microencapsulated iron added to cereal to be in the range of 2–8%. Thus, the total amount of available iron in an 80-mg sachet is 1.6–6.4 mg.

There is little recent data on iron absorption in infants receiving ferrous sulfate drops. On the basis of a compilation of older data, we assumed that iron absorption would be in the range of 4–18%, making the total amount of available iron 1.6–7.2 mg/d (13–15). Thus, the range of potentially absorbed iron was similar, but not identical, for the 2 forms of iron. Because the ferrous fumarate was coated, we anticipated that there would be minimal intestinal irritation from the relatively high 80-mg dose of iron.

During the baseline assessment, a written questionnaire was administered to collect demographic, nutritional, and health data for each infant. Field workers visited infants at 2-wk intervals after the baseline visit, for a total of 5 visits. At each visit, a questionnaire about side effects, ease of use, and compliance over the preceding 7 d was completed. Side effects included the incidence of diarrhea, constipation, and general discomfort after ingestion of the iron drops. Questions about ease of use included whether the children objected to taking the iron and whether microencapsulated ferrous fumarate changed the color, taste, or texture of the infants' food. To evaluate compliance during each visit, the number of used (empty) sachets was counted or unused drops measured. At each visit, fieldworkers provided parents with oral educational reinforcement to maximize compliance with the intervention.

Anthropometric measurements were completed during the baseline and final visits. An infant-length board with a sliding foot board was used to measure the child's body length, and a hanging scale graduated in 100-g divisions was used for weight measurements. Two fieldworkers completed the measurements in duplicate using standardized WHO techniques (16).

Capillary blood samples taken at the baseline and final visits were obtained from a finger prick with the use of aseptic techniques; hemoglobin concentrations were measured immediately with a portable HEMOCUE B-hemoglobin photometer (Hemocue Inc, Angelholm, Sweden) by trained technicians using standardized techniques (17). Malaria parasite smears were taken (at the baseline visit only) and 500- μ L blood samples were collected and preserved in ice-lined cold boxes. Blood samples were returned to the base station within 6 h of collection, where the serum was separated by centrifugation (12000 \times g, 10 min, room temperature) before storage at -40° C. Serum ferritin was assayed in duplicate with a commercial enzyme-linked immunosorbent assay (Spectro Ferritin Kit; Ramco Laboratories, Houston) (18). Baseline and end-of-treatment ferritin samples from an individual subject were assayed on the same day (in a single batch) on one 96-well microtiter plate to minimize interassay variation. An external reference standard (Lyphochek Anemia Control; Bio-Rad, Anaheim, CA) was assayed in duplicate on each microtiter plate for the ferritin assay.

Sample size and power

The primary outcome was the successful treatment of anemia (ie, the proportion of children with hemoglobin concentrations

≥ 100 g/L). On the assumption of 90% cure rates in the drops group and 80% in the sprinkles group and with a type I error set at 0.05 and a 0.9 probability of detecting a true difference, the final sample size estimate was 286 subjects per group. It was our expectation that 65% of the infants in the Kintampo region would have hemoglobin values within our target range (70–99 g/L); therefore, a total of 880 infants were screened to obtain the needed number of subjects for the study.

Data processing and analysis

Data forms were manually checked for completeness and consistency before submission for processing. Data were entered twice by 2 different data-entry clerks in VISUAL FOX PRO 6.0 (Microsoft Corporation), verified, and checked for range and consistency with customized data-entry and processing programs (Microsoft Access 97; Microsoft Corporation). Data queries were forwarded to the Kintampo field office and resolved, whenever possible, by rechecking original data forms or were verified in a repeat home visit if indicated. Monthly summary reports, including the entered data, were sent electronically from Kintampo to the Central Study Centre in Toronto via the Internet and a file-transfer-protocol host site.

Data were analyzed with SAS software (version 6.12; SAS Institute, Inc, Cary, NC). The proportion of children who were successfully treated were compared between the groups with chi-square analysis. Paired *t* tests were used to analyze the change in hemoglobin, ferritin, and anthropometric measurements over time. Differences between groups in hemoglobin, ferritin, and anthropometric measurements from the beginning to the end of the study were assessed with Student's *t* test and the interaction between treatment and time of the study was assessed with use of a two-factor, repeated-measures analysis of variance (with PROC GLM in SAS version 6.12). All analyses of ferritin concentrations were conducted on log-transformed data because of their skewed frequency distribution. The acceptable level of statistical significance for all tests was $P < 0.05$. Results are expressed as geometric means (\pm SDs) and ranges.

RESULTS

After the screening survey, 557 infants with hemoglobin concentrations between 70.0 and 99.9 g/L were randomly assigned to treatment. Sixty-four (11.5%) of the 557 infants did not attend the final assessment visit. This loss was similarly distributed between the 2 groups; moreover, there were no significant differences in baseline characteristics between these infants and the infants who successfully completed the trial. Consequently, a total of 493 infants completed the final assessment, including anthropometric measurements and blood sampling.

There were no significant differences in age (12.9 ± 4.5 mo) or in hemoglobin or ferritin concentrations (Table 1) between the 2 treatment groups at baseline. At baseline, the infants in both groups were stunted and wasted: their weight-for-age, height-for-age, and weight-for-height *z* scores were all < 0 . Stunting, as defined by Waterlow's (19) criterion of a height-for-age *z* score < -2 , was found in 150 (30.4%) of the 493 subjects at the final assessment.

Despite random allocation to treatment, there was an imbalance in the distribution of sexes in the treatment groups: boys were underrepresented in the intervention group [95 of 245 (39%) compared with 139 of 247 (56%) in the drops group]. However, hemo-

TABLE 1
Hemoglobin and ferritin concentrations and the percentage of anemic subjects, by treatment group, at baseline and after 2 mo of treatment¹

	Sprinkles group	Drops group
Baseline		
Hemoglobin (g/L) ²	87 \pm 8 [246]	87 \pm 9 [247]
Anemic subjects (%) ³	100	100
Ferritin (μ g/L) ^{4,5}	38.8 (0.34–345.7) [212]	40.0 (0.16–366.4) [217]
End of treatment		
Hemoglobin ²	102 \pm 16 [246] ⁶	100 \pm 17 [247]
Anemic subjects (%) ³	42.3	43.7
Ferritin (μ g/L) ^{4,5}	71.2 ⁶ (0.70–397.3) [212]	106.8 ^{6,7} (6.58–391.8) [217]

¹*n* values in brackets; ranges in parentheses.

² $\bar{x} \pm$ SD.

³Anemia is defined as a hemoglobin concentration < 100 g/L.

⁴Geometric \bar{x} .

⁵The analysis was done with log-transformed values because concentrations were not normally distributed.

⁶Significantly different from baseline, $P < 0.001$.

⁷Significantly different from the sprinkles group, $P = 0.001$.

globin values were not correlated with sex at baseline or at 2 mo. There were no significant differences in the prevalence of positive malaria smears [158 of 237 (66.2%) in the sprinkles group and 160 of 240 (66.7%) in the drops group] between the 2 groups.

Hemoglobin

In both groups, there was a significant increase in hemoglobin concentrations from baseline to the end of treatment study ($P < 0.001$; Table 1). The change in hemoglobin concentrations was not significantly different between groups. Fifty-seven percent (281 of 493) of infants advanced from an anemic to a nonanemic state (hemoglobin ≥ 100 g/L). This rate was not significantly different between groups: 57.7% (142 of 246 subjects) in the sprinkles group and 56.3% (139 of 247 subjects) in the drops group. The relative risk of remaining anemic after 2 mo of treatment was 1.03 times greater in the drops group than in the sprinkles group, but the difference was not significant.

Data were also analyzed to determine the percentage of infants who positively responded to iron treatment; a positive response was defined as an increase in hemoglobin of ≥ 10 g/L in the final blood sample). In the sprinkles group, 166 of 246 (67.5%) of the children responded; in the drops group, 150 of 247 (60.7%) responded. There was no significant difference between treatment groups.

At baseline, as expected, there was a significant difference in hemoglobin concentrations between malaria-positive and -negative infants (85.6 ± 8.4 compared with 89.5 ± 7.7 g/L, respectively; $P < 0.0001$). Independent of group designation, infants who tested positive for malaria were more likely to be anemic at the end of 2 mo. The relative risk of remaining anemic after 2 mo of treatment was 1.23 times greater in those with malaria than in those who were malaria-free (95% CI: 1.10, 1.37; $P = 0.0006$).

Ferritin

Baseline ferritin concentrations were similar between treatment groups (Table 1). There was a significant increase in both groups after 2 mo of treatment; however, the mean value was significantly higher in the drops group than in the intervention sprinkles group.

This is reflected in the significant treatment group \times time interaction. The range of ferritin concentrations was wide at both baseline and at the end of treatment, as is usual with the wide interindividual and analytic variance associated with this measure, especially in a region in which malaria is endemic (20, 21).

Anthropometric measurements

No treatment effect was found for weight-for-age or weight-for-height z scores in either group. There was a significant decrease in height-for-age z scores in both groups from baseline to the final measurements (change: -0.16 ± 0.65 ; $P < 0.001$).

Side effects and adherence to treatment

Starting 2 wk after baseline and then every 2 wk for the duration of the study, mothers were asked about compliance with treatment over the preceding 7 d, and fieldworkers checked supplies of drops and sachets. On the basis of combined data from the 4 monitoring visits, 92% of the drops group received the drops on ≥ 4 d/wk, whereas 81% never missed a day. Only 1.7% of the group took no drops during the preceding week. Eighty-three percent of the children in the sprinkles group complied with treatment on ≥ 4 d/wk, 66% never missed a day, and 5.5% received no sprinkles during the preceding 7 d.

Seventy-four percent (933 of 1277) of the mothers of children in the drops group reported that their children objected to taking the drops in some way (eg, they cried, made a funny face, and tried to keep their mouth shut when the drops were administered). Sixteen percent of the mothers of children in the sprinkles group also reported having problems giving their children sprinkles (eg, children did not want to eat the food into which the sprinkles were added). Their mothers did not perceive that the sprinkles changed the color or texture of the food to which they were added. Reported side effects were rare and mild and consisted mainly of diarrhea. There were no significant differences in side effects between the groups. Diarrhea was reported in 76 of 523 (14.5%) subjects in the drops group and in 62 of 486 (12.8%) subjects in the sprinkles group.

DISCUSSION

Use of sprinkles or drops containing iron resulted in similar increases in hemoglobin from the beginning to the end of treatment and in similar rates of successful treatment of anemia. To our knowledge, this is the first time microencapsulated iron was used for the treatment of anemia.

Although we did not use a placebo control group for ethical reasons, it is unlikely that the observed improvement in anemia would have occurred had no treatment been provided. The typical complementary weaning food in Ghana, a maize-based porridge, has a low iron content (< 1.3 mg Fe per 100 g cereal) and poor iron bioavailability (12). In addition, malaria and parasite infections are common in Ghana. Consequently, children with moderately severe anemia would likely have remained anemic if treatment were not provided.

After the 2-mo intervention, $\approx 40\%$ of infants remained anemic. There are many possible explanations for this observation. This study took place during the wet season (from May to August); thus, not surprisingly, 65% of infants in the study tested positive for malaria at baseline. Malaria has been shown to be a significant contributor to the etiology of anemia, especially in young infants in highly endemic areas (22). Parasitic and *Helicobacter pylori* infections, which are common in West Africa,

may also contribute to continuing blood losses and bone marrow unresponsiveness (23). Other endemic infections, such as gastroenteritis and respiratory infections, might also interfere with the utilization of absorbed iron. Another possible reason for the lack of response to iron was a subclinical vitamin A deficiency because infants in Ghana are known to be at risk of this deficiency (24). Previous epidemiologic studies indicated that vitamin A deficiency and anemia often coexist (25). It is likely that vitamin A plays a role in mobilizing iron from stores to be used for hematopoiesis (26). Thus, it is possible that those infants who did not respond to the drops or sprinkles did not have iron deficiency anemia but had other causes of anemia.


At the end of the 2 mo of treatment with iron, serum ferritin concentrations (an indirect measure of iron stores) had increased significantly in both groups. These results suggest that more iron was absorbed than was needed for immediate erythropoiesis. However, the changes in ferritin concentrations from baseline to the end of the study were greater in the drops group than in the sprinkles group. Despite the presence of ascorbic acid and the higher total dose of iron in the sprinkles group, the bioavailability of iron was likely lower from the sprinkles than from the drops. This observation is not surprising because the sprinkles were mixed with food, which would likely impede iron absorption more than would drops that were taken without food. Increasing the dose per sachet of sprinkles would likely offset this difference; however, this was not necessary for the treatment of anemia. The only theoretical advantage of increasing the dose per sachet would be an even greater increase in iron stores above those observed in the current study, with the possible disadvantage of gastrointestinal irritation.

One might argue that iron absorption would be enhanced if the iron dose in the sprinkles were divided among 2–3 servings/d because fractional iron absorption is inversely related to the dose (27). However, we believe that compliance would be compromised as a result of such a regimen and that this disadvantage would outweigh the advantages. In a nonstudy setting, we intuitively expect greater compliance with once-daily sprinkles.

Our findings suggest important policy and program implications for the treatment of anemia. Although we did not measure compliance with the different dosing regimens directly (the protocol was not designed as an effectiveness study), our data suggest that compliance was sufficient to treat anemia and increase iron stores. Compliance with the sprinkles (67% of the group never missed a dose) was less than it was with the drops. There are many likely explanations for this difference. Many infants in the study who had just reached 6 mo of age at baseline were just starting to consume complementary foods (in addition to breastfeeding). Some of the infants at this age were not yet consuming these foods daily. Because sprinkles had to be added to complementary foods, if these foods were not eaten the sprinkles would not be provided. In addition, during the study there was a 2–4-wk period during which the sprinkles were difficult to use because humidity had caused them to turn gummy inside the sachet. Many mothers noted that the gummy sprinkles were difficult to remove from the sachet and, thus, they did not sprinkle them onto the food. This problem was solved by changing the packaging from 2 layers (paper and polyethylene) to 3 layers, consisting of paper, polyethylene, and foil halfway through the study. In fact, by the final visit, compliance with treatment was not significantly different between the groups. Additional appropriately

designed research is needed to determine the likely effectiveness of the sprinkles in a nonstudy setting.

We believe that sprinkles will be widely accepted and preferable to iron drops as a means of iron supplementation if they are promoted in this population. Sprinkles added to food do not change the color or texture of the food. The taste of infant cereals is not affected by the addition of sprinkles, at least from an adult perspective. A single-dose sachet is simple to use and sprinkles are less expensive to manufacture and distribute than are drops. Finally, the likelihood of an accidental overdose from the ingestion of too much iron in sprinkles is negligible because numerous individual packages would have to be opened and ingested for this to occur. A further advantage of the sprinkles is that more than one micronutrient can be included in the sachet, depending on the needs of a specific locale. For example, iron, ascorbic acid, vitamin A, zinc, and iodine can easily be combined and included in the sachet. From a practical perspective, the option of using sprinkles, therefore, may improve compliance with treatment and thus the success rate for the treatment of anemia.

The results of this study indicate that, in a controlled setting, micronutrient sprinkles result in a rate of successful treatment of anemia comparable with the current standard form—ferrous sulfate drops. Further studies testing the effectiveness of sprinkles for the treatment and prevention of anemia are warranted. 

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