Dual fortification of salt with iodine and micronized ferric pyrophosphate: a randomized, double-blind, controlled trial[1–3]

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ABSTRACT

Background: In many developing countries, children are at high risk for both goiter and anemia. In areas of subsistence farming in rural Africa, salt is one of the few regularly purchased food items and could be a good fortification vehicle for iodine and iron, provided that a stable yet bioavailable iron fortificant is used.

Objective: We tested the efficacy of salt dual-fortified with iodine and micronized ferric pyrophosphate for reducing the prevalence of iodine and iron deficiencies in children.

Design: In rural northern Morocco, we fortified local salt with 25 μg I (as potassium iodate)/g salt and 2 mg Fe (as micronized ferric pyrophosphate; mean particle size = 2.5 μm)/g salt. After storage and acceptability trials, we compared the efficacy of the dual-fortified salt (DFS) with that of iodized salt in a 10-mo, randomized, double-blind trial in iodine-deficient 6–15-y-old children ($n = 158$) with a high prevalence of anemia.

Results: After storage for 6 mo, there were no significant differences in iodine content or color lightness between the DFS and iodized salt. During the efficacy trial, the DFS provided ≈18 mg Fe/d; iron absorption was estimated to be ≈2%. After 10 mo of treatment in the DFS group, mean hemoglobin increased by 16 g/L ($P < 0.01$), iron status and body iron stores increased significantly ($P < 0.01$), and the prevalence of iron deficiency anemia decreased from 30% at baseline to 5% ($P < 0.001$). In both groups, urinary iodine ($P < 0.001$) and thyroid volume ($P < 0.01$) improved significantly from baseline.

Conclusion: A DFS containing iodine and micronized ferric pyrophosphate can be an effective fortification strategy in rural Africa. *Am J Clin Nutr* 2004;80:952–9.

KEY WORDS Iodine, iron, micronized ferric pyrophosphate, iodine deficiency, iron deficiency, dual fortification of salt, anemia, goiter, children

INTRODUCTION

Iodine deficiency disorders and iron deficiency anemia (IDA) affect ≥30% of the global population (1, 2). These deficiencies often coexist—in regions of West and North Africa, 20–38% of schoolchildren may suffer from both goiter and IDA (3, 4). IDA impairs thyroid metabolism and reduces the efficacy of iodine prophylaxis in areas of endemic goiter (3, 5–7). Although universal salt iodization has proven highly effective against iodine deficiency disorders (2), effective iron fortification of foods remains a challenge (8, 9). Salt is a good potential vehicle for iron and iodine, particularly in rural West and North Africa, because in poor regions of subsistence farming, salt is one of the few regularly purchased food items (4, 10). In these regions, dual fortification of salt with iodine and iron could be an effective strategy.

However, maintaining iodine stability and iron bioavailability while avoiding color change in dual-fortified salt (DFS) is difficult, particularly in low-grade salt in developing countries (11–14). Water-soluble, highly bioavailable iron compounds react with moisture and impurities in salt, which causes color changes and iodine losses (14). We recently showed the efficacy of a DFS containing ferrous sulfate encapsulated with partially hydrogenated vegetable oil (4). However, despite encapsulation, the ferrous sulfate produced a yellow color change in the DFS when salt moisture content was high (4). Currently available forms of encapsulated ferrous sulfate and ferrous fumarate cause unacceptable color changes when added to low-grade salt in Africa (15).

Poorly soluble iron compounds, such as elemental iron powders or iron phosphates, tend to cause fewer sensory changes in foods (8, 9). Ferric pyrophosphate (FePP) has a white color and produces negligible color change when added to local salt in West and North Africa (15). When FePP is added to salt containing potassium iodate (KIO_3), iodine stability is comparable to that in iodized salt (IS) (15). Although most commercial forms of FePP have a relative bioavailability (RBV) ≤ 50% of ferrous sulfate (8, 9), reducing the particle size of FePP increases its absorption. The absorption of FePP with a mean particle size (MPS) of ≈0.5 μm is comparable with that of ferrous sulfate (16). In rats, the RBV of FePP with an MPS of 2.5 μm is ≈70%

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of that of ferrous sulfate (R Wegmüller, personal communication, 2004). Micronized FePP may therefore have excellent potential as an iron fortificant in DFS.

We formulated a DFS containing potassium iodate and micronized FePP and tested its stability in local salt and its organoleptic qualities when added to local meals. We then compared the efficacy of the DFS with that of IS in a randomized, double-blind trial in iodine-deficient Moroccan children with a high prevalence of anemia.

SUBJECTS AND METHODS

The study was conducted in villages in the Brikcha Rural Commune, an area of endemic goiter in northern Morocco (17). The region is 400–800 m above sea level, and the climate is temperate, with an 8-mo dry season (22–34 °C, mean rainfall of 23 cm/mo) and a 4-mo damp season (10–22 °C, mean rainfall of 77 cm/mo). The population is of mixed Berber and Arab descent. This region is isolated from commercial routes, >95% of the population is rural, and most available food is produced locally on small farms (18). Per capita salt and iron intakes in school-age children in this region are 7–12 g/d and 9–14 mg/d, respectively (4). Iron bioavailability from the local diet is estimated to be 2–4% (4) when adjusted for low body iron stores (19).

Salt preparation

A local cooperative supplies nearly all salt in this region; the salt is produced in drying ponds by using water from a salty spring. The salt (95.4% NaCl) is not washed or ground and has a milky-white color and an average crystal size of 1.4 mm. Its moisture content is <1% during the dry season but 3–4% during the damp season, and it contains 2.5% $CaSO_4$, <0.1% $MgSO_4$, and <2 parts per million iodine. Morocco legislated mandatory salt iodization in 1997; it is estimated that ≈45% of the population has access to IS (N Chaouki, personal communication, 2002). Because of financial constraints, this small local cooperative has not yet begun iodization. To prepare the IS and DFS, iodine was added as reagent-grade potassium iodate (Sigma & Aldrich, Buchs, Switzerland) at a concentration of 25 μg iodine/g salt. The DFS was fortified with food-grade micronized FePP (art. no. 3043448; Paul Lohmann AG, Emmerthal, Germany). This FePP contains 21% iron and has an MPS (d_{50}) of 2.5 μm (d_{10}, 0.4 μm; d_{90}, 6.1 μm). On the basis of mean local salt intakes of 7–12 g/d in children (4), iron bioavailability of 2–4% from the diet (4), and an RBV of 70% for the micronized FePP, we chose a fortification concentration of 2 mg Fe/g salt for the DFS. Our fortification goal was to provide ≥0.5 mg absorbed Fe/d to the children in the study. Concentrated premixes were made by adding 0.84 g KIO_3 (for the IS) or 0.84 g KIO_3 and 190 g FePP (for the DFS) to 2-kg batches of salt by using a small, electric, rotating-drum mixer (MINI 80; Engelsmann, Ludwigshafen, Germany) at 26 rpm for 10 min. The 2-kg premixes were then mixed into 18-kg batches of salt in a large, electric, rotating-drum mixer (ELTE 650, Engelsmann) at 30 rpm for 10 min.

Stability and acceptability testing

The IS and DFS were locally stored as 2-kg batches in closed, low-density, transparent polyethylene bags that were kept indoors and out of direct sunlight. After storage for 0, 2, 4, and 6 mo, 30-g salt aliquots ($n = 3$) were taken for measurement of iodine concentration in the IS and of iodine and iron concentrations in the DFS. A 6-mo study was done to approximate the time required for the production, distribution, and consumption of salt in this region. Color stability was determined by reflectance colorimetry as well as panel visual inspection by 6 local adults of unmarked samples side-by-side on white backgrounds. To judge DFS and IS acceptability after 10 mo of household salt use during the efficacy study, an interview was conducted in 50% of randomly selected households in the IS ($n = 31$) and DFS groups ($n = 25$). The head of the household, who was blind to group assignment, answered forced-choice questions on patterns of salt use, color and taste acceptability, and overall satisfaction with the salt.

Organoleptic testing

To evaluate potential sensory changes in local foods, IS and DFS were added to meals that were prepared by local women in their kitchens with the use of traditional recipes. Each type of salt was added in identical amounts in parallel to separate portions of 4 common foods: bread (made with mainly white flour), bisarra (fava bean and olive oil purée), chaaria (semolina noodles in milk), and couscous (semolina). The flavor, odor, and color of these foods were then compared by a panel of 18 local adults ($\bar{x}$ age: 33 y; 59% female) with the use of triangle tests (20). During the triangle test, 3 coded samples of each of the 4 foods were given in random order in a private setting. The panelists were to determine which sample differed from the other 2 samples and describe how they differed (20).

Efficacy study

The subjects were 6–15-y-old children from 2 neighboring primary schools. Informed written consent was obtained from the chief medical officer and school directors, and informed oral consent was obtained from the parents of the children. The Swiss Federal Institute of Technology in Zürich, Switzerland, and the Ministry of Health in Rabat, Morocco, gave ethical approval for the study. All children from the schools were invited to participate in the 10-mo study, and all accepted ($n = 163$) and were enrolled. At baseline, weight and height were measured, and a casual spot urine sample was collected for measurement of urinary iodine (UI) concentration. Five milliliters of whole blood was collected by venipuncture for determination of hemoglobin, C-reactive protein (CRP), serum ferritin (SF), whole-blood zinc protoporphyrin (ZnPP), and serum transferrin receptor (TfR). Thyroid gland volume (T_{vol}) was measured by using a portable Aloka SSD-500 Echocamera (Aloka, Mure, Japan) with a high-resolution, 7.5-MHz linear transducer (21).

Because each participating family shared a monthly salt portion, children were randomly assigned at the household level into 2 groups: one group ($n = 86$) was given the IS and the second group ($n = 77$) was given the DFS. Both investigators and households were blind to group assignment. The IS and the DFS were prepared as described above. For monitoring, 30-g aliquots ($n = 6$) of the IS and the DFS were taken and measured for iodine and iron content at each monthly mixing. On the basis of a per capita salt intake of 12 g/d and local census data indicating an average of 7.5 members per household, each household was provided 2 kg salt to supply all household needs at the beginning of each month for 10 mo. The salt was dispensed directly to the head of the household from a central supply at the local health center. At

baseline, the study was carefully explained to the participating families, and it was emphasized that the new salt should be used for all cooking and food preparation, as well as at the table. This message was reinforced at each of the monthly salt distributions.

At 5 and 10 mo, the weight and height of the children were measured, and casual spot urine samples were collected for measurement of UI. Whole blood was collected by venipuncture for determination of hemoglobin, CRP, SF, ZnPP, and TfR. T_{vol} was measured with the use of ultrasound. After completion of the fortification trial, all remaining children with IDA were treated with oral iron (60 mg Fe as ferrous sulfate 4 d/wk for 12 wk).

Laboratory analyses

Serum and urine samples were separated into aliquots and frozen at −20 °C until analysis. UI was measured by using the Pino modification of the Sandell-Kolthoff reaction (22). At UI concentrations of 47 and 79 μg/L, the interrun CV of this assay in our laboratory was 10.3% and 12.7%, respectively. The limit of detection was 2 μg/L; samples below this limit were assigned a value of 0. T_{vol} was calculated by using the method of Brunn et al (23). MBZ performed all ultrasound measurements during the study. To estimate intraobserver variability, duplicate T_{vol} measurements were taken in 20 children at the 5- and 10-mo time points; the mean (±SD) intraobserver variability was 3.9 ± 2.1%. Goiter was defined by using sex- and body surface area–specific reference criteria for T_{vol} (21).

Hemoglobin was measured by using an AcT8 Counter (Beckman Coulter, Krefeld, Germany). Anemia was defined as a hemoglobin concentration < 130 g/L in boys aged ≥15 y, a hemoglobin concentration < 120 g/L in children aged ≥12 y and in girls aged ≥15 y, and a hemoglobin concentration < 115 g/L in children aged 5–11 y (24). ZnPP was measured on washed red blood cells by using a hematofluorometer (Aviv Biomedical, Lakewood, NJ). SF and TfR were measured by using enzyme-linked immunosorbent assays (RAMCO, Houston). CRP was measured by using nephelometry (TURBOX; Orion Diagnostica, Espoo, Finland). Normal reference values are as follows: ZnPP, ≤40 μmol/mol heme; SF, 15–300 μg/L; TfR, 2.9–8.5 mg/L; CRP, <10 mg/L. Iron deficiency was defined as either an SF concentration < 15 μg/L or as a TfR concentration > 8.5 mg/L plus a ZnPP concentration > 40 μmol/mol heme (24, 25). Body iron was estimated by using the method of Cook et al (26) with the following formula:

$$\text{Body iron (mg/kg)} = -[\log(\text{TfR/SF}) - 2.8229]/0.1207 \quad (1)$$

To measure salt color, 10 g salt was transferred into a glass container for color determination on the Hunter scale with the use of a Spectral Photometer (Chroma Meter CR-310; Minolta, Osaka, Japan) with illuminant D_{65} (average daylight, including ultraviolet spectra), a 0° observer angle, and a large reflectance spectrum. On the Hunter scale, the *L* value measures light reflection (a value of 100 is pure white; a value of 0 is pure black), the *a* value is a measure of redness and greenness, and the *b* value is a measure of yellowness and blueness. Each sample was measured 3 times, and the glass container was rotated 90° after each measurement. The color of the DFS was then compared with that of the IS, and the absolute color difference was expressed by Δ*E* (15). Δ*E* was calculated by using the following equation:

$$\Delta E = \sqrt{(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2} \quad (2)$$

where Δ*L*, Δ*a*, and Δ*b* describe the difference between the color of the DFS and the reference color (IS).

For DFS, determination of iodine content by titration with thiosulfate (27) is complicated by interference from iron in the salt. We therefore measured iodine in salt aliquots dissolved in distilled water by using a modification of the Sandell-Kolthoff reaction (15, 28). At iodine concentrations of 30 μg/g salt, the interrun CV of this assay in our laboratory was 7%. This method shows good agreement ($r^2 = 0.97$) with isotope abundance ratio measurements by mass spectrometry (28). The iron content of the salt was analyzed by using a standard addition method with flame atomic absorption spectroscopy (SpectrAA-400; Varian, Mulgrave, Australia). The mineral composition of the native salt was analyzed by using inductively coupled plasma optical emission spectrometry (Spectro Modula; Spectro Analytic Instruments GmbH and Co KG, Kleve, Germany), and the particle size distribution was measured by sieve analysis.

Statistical analyses

Data processing and statistical analyses were performed with the use of SPLUS (2000; Insightful Corporation, Seattle), PRISM (version 3; GraphPad, San Diego), and EXCEL (XP 2002; Microsoft, Seattle). Two-factor repeated-measures analysis of variance was done to compare the effects of group × time for hemoglobin, CRP, SF, TfR, ZnPP, body iron, T_{vol}, UI, and salt color and iodine content. If the interaction effect was significant ($P < 0.05$), *t* tests between groups and paired *t* tests within groups were done and adjusted for multiple comparisons (Bonferroni correction). Proportions were compared by using the chi-square test. When data were not normally distributed, statistical analysis was done after log transformation. Logistic regression was done to compare effects of group × time for the binary variables of anemia, IDA, iron deficiency without anemia, and goiter. Significance was set at $P < 0.05$.

RESULTS

Stability

The color stability, iodine content, and iron content of the IS and the DFS at 0, 2, 4, and 6 mo after mixing are shown in **Table 1**. By colorimetry, there was no significant difference between the IS and the DFS in the *L* color value, and no significant color change in the IS or the DFS over 6 mo of storage. In comparing the DFS with the IS, the Δ*E* of 6–7 for the DFS was due to the slight difference in color shade between the DFS (light beige) and the IS (milky white). There was no significant difference between the DFS and the IS in iodine content during storage; both salts lost ≈20% of their iodine content after 6 mo.

Acceptability and organoleptic testing

The results of interviews conducted after 10 mo of household salt use are shown in **Table 2**. Both salts were universally used, and there were no significant differences between the DFS and the IS in acceptability of salt color, salt taste in foods, and overall acceptability. Sixteen percent and 24% of the IS and the DFS heads of household, respectively, noted a mild color change in

TABLE 1
Comparison of color and iodine concentration in the iodized salt (IS) and the dual-fortified salt (DFS) containing iodine and iron and of iron concentration in the DFS at mixing (0 mo) and after 2, 4, and 6 mo of storage[1]

	Color						
	Lightness[2]		ΔE[3,4]		Iodine[5]		
Length of storage (mo)	IS	DFS	IS	DFS	IS	DFS	Iron (DFS)
					μg/g salt		*mg/g salt*
0	80.9 ± 0.88	81.9 ± 0.10	0.88 ± 0.09	6.77 ± 0.15	27.4 ± 3.8	24.2 ± 9.4	2.02 ± 0.17
2	80.3 ± 0.45	81.9 ± 0.27	1.28 ± 0.43	6.94 ± 0.15	18.6 ± 2.1	20.7 ± 3.8	2.26 ± 0.38
4	80.7 ± 1.01	79.5 ± 0.10	0.96 ± 0.09	7.13 ± 0.32	21.0 ± 3.1	20.2 ± 2.6	1.97 ± 0.23
6	80.3 ± 0.75	79.5 ± 0.50	1.34 ± 0.73	7.16 ± 0.54	19.4 ± 1.4	20.1 ± 2.7	2.25 ± 0.07

[1] All values are $\bar{x}$ ± SD; $n = 3$ for the IS and 3 for the DFS at all time points.
[2] Lightness scale: 1 = black, 100 = white.
[3] ΔE = absolute color difference between DFS or IS and an IS reference sample.
[4] Significant main effect of fortification, $P < 0.001$.
[5] Significant main effect of time, $P < 0.01$.

one or more foods (mainly egg and milk dishes) when the respective salts were added. However, in the triangle testing that compared the IS to the DFS, there was no significant difference in color, odor, or taste between the salts in any of the traditional foods. Three of the foods tested—bread, chaaria, and couscous—were pale colored and mild tasting.

Efficacy trial

The characteristics of the treatment and control groups at baseline are shown in **Table 3**. Randomization at the household level was effective; there were no significant differences in baseline characteristics between the groups. None of the children had severe anemia (hemoglobin concentration < 80 g/L) at baseline. Of the 163 children who began the study, 158 completed it; 5 children moved away or were absent from school on the measurement days (3 in the IS group and 2 in the DFS group). In the monitoring aliquots of the IS and the DFS taken at the monthly mixings, the mean (±SD) iodine concentrations in the IS and the DFS were 22.8 ± 5.1 and 20.4 ± 6.4 μg/g salt, respectively (NS).

TABLE 2
Acceptability of iodized salt (IS) and dual-fortified salt (DFS) containing iodine and iron after 10 mo of salt use[1]

	Salt	
Question	IS	DFS
	%	
Salt quantity used monthly in the household?		
≤1 kg	0	0
1–2 kg	24	32[2]
2 kg[3]	52	42[2]
>2 kg	24	26
Salt consumed every day by children?	100	100
Salt used for all foods during cooking and at table?	100	100
Salt changed the color of foods?	16	24[2]
Salt color acceptable in damp and dry seasons?	100	98
Salt taste acceptable in all foods?	96	100
Salt acceptable overall?	100	100

[1] Percentages of positive answers are shown.
[2] Significantly different from IS, $P < 0.05$.
[3] Amount distributed monthly to each study household.

The mean iron concentration in the DFS at the monthly mixings was 2.17 ± 0.77 mg/g salt.

Changes in hemoglobin, iron status indexes, and body iron are shown in **Table 4**. In comparison with the mean hemoglobin concentrations in the IS group, mean concentrations in the DFS group increased significantly from baseline at 5 and 10 mo ($P < 0.01$). All indexes of iron status (SF, TfR, and ZnPP) and body iron stores improved significantly in the DFS group compared with the IS group at 5 and 10 mo ($P < 0.01$) (Table 4). After 10 mo, mean (±SD) total body iron increased in the DFS group from 39.8 ± 74.2 to 145.7 ± 81.4 mg, whereas mean total body iron decreased in the IS group from 44.7 ± 89.2 to 35.1 ± 90.7 mg. There were no significant differences between the groups in mean CRP concentration or the prevalence of elevated CRP at 0, 5, and 10 mo; 4.1–6.4% of the children had elevated CRP (data not shown). As shown in **Figure 1**, the prevalences of anemia, IDA, and iron deficiency without anemia were sharply lower in the DFS group than in the IS group at 5 and 10 mo ($P < 0.001$).

There were no significant differences in median UI concentration between the 2 groups throughout the study (**Table 5**). In both groups, median UI concentrations increased significantly from baseline at 5 and 10 mo ($P < 0.001$). At 10 mo, median UI had increased to near the cutoff value (100 μg/L) for risk of

TABLE 3
Characteristics of the children in the iodized salt (IS) and dual-fortified salt (DFS) groups at baseline[1]

Characteristic	IS group (n = 40 F, 43 M)	DFS group (n = 35 F, 40 M)
Age (y)	10.6 ± 2.4[2]	10.8 ± 2.3
Weight (kg)	31.2 ± 9.9	29.8 ± 8.3
Height (m)	1.34 ± 0.14	1.36 ± 0.14
Prevalence of anemia [n (%)]	55 (46)	60 (45)
Prevalence of iron deficiency anemia [n (%)]	34 (28)	31 (23)
Prevalence of iron deficiency without anemia [n (%)]	55 (46)	52 (39)

[1] There were no significant differences between the 2 groups.
[2] $\bar{x}$ ± SD (all such values).

TABLE 4
Hemoglobin transferrin receptor (TfR), zinc protoporphyrin (ZnPP), serum ferritin (SF), and body iron concentrations in the iodized salt (IS) and dual-fortified salt (DFS) groups over 10 mo[1]

Time (mo)	Hemoglobin		TfR		ZnPP		SF		Body iron	
	IS group	DFS group	IS group	DFS group	IS group	DFS group	IS group	DFS group	IS group	DFS group
	g/L		*mg/L*		*μmol/mol heme*		*μg/L*		*mg/kg*	
0	116 ± 9[2]	113 ± 10	7.5 ± 2.4	7.7 ± 3.0	50 ± 36	49 ± 32	15.2 (7.1, 25.9)[3]	15.5 (6.4, 28.1)	1.23 ± 2.64	1.26 ± 2.36
5	116 ± 11	126 ± 10[4,5]	7.0 ± 1.9[6]	6.1 ± 1.7[4,7]	49 ± 37	32 ± 24[5,6]	19.0 (7.3, 38.2)	29.8 (13.3, 78.5)[4,5]	2.21 ± 2.79[6]	4.34 ± 2.48[4,5]
10	115 ± 8	128 ± 11[4,5]	7.7 ± 2.4	5.8 ± 1.1[4,5]	52 ± 35	27 ± 20[4,5]	15.0 (6.9, 28.1)	33.1 (12.5, 76.4)[4,5]	1.08 ± 3.10	4.82 ± 1.94[4,5]

[1] Significant treatment × time interaction for all variables, $P < 0.001$ (ANOVA).
[2] $\bar{x} \pm$ SD (all such values).
[3] Geometric $\bar{x}$; −1 SD and +1 SD in parentheses.
[4,6] Significantly different from baseline: [4]$P < 0.01$, [6]$P < 0.05$.
[5,7] Significantly different from the IS group: [5]$P < 0.01$, [7]$P < 0.05$.

iodine deficiency from the World Health Organization/International Council for the Control of Iodine Deficiency Disorders (1). In both groups, mean T_{vol} decreased significantly from baseline at 5 and 10 mo ($P < 0.01$), and there was a significant decrease in goiter prevalence ($P < 0.01$). However, at 10 mo, the mean percentage change in T_{vol} from baseline was significantly higher in the DFS group than in the IS group ($P < 0.01$), and goiter prevalence was also significantly lower ($P < 0.01$) in the DFS group at 10 mo.

DISCUSSION

Previous attempts to develop an effective DFS produced mixed results. In India, Mannar et al (29) reported that the iodine content and color of a DFS containing potassium iodide and ferrous fumarate was stable for 8 wk in waterproof packaging. The Indian National Institute of Nutrition (NIN) formulated a DFS containing ferrous sulfate, potassium iodide, and a stabilizer, sodium hexametaphosphate (11, 13). Mean iron absorption from the NIN DFS consumed with rice was 6.1%, and the addition of sodium hexametaphosphate increased iron absorption by ≈50% (30). The stability of iodine in the NIN DFS depended on salt quality; in the presence of magnesium chloride as an impurity, the salt lost significant iodine (31). In field trials of the NIN DFS, one study found no overall benefit on hemoglobin concentrations, whereas in a second study, hemoglobin concentrations decreased significantly in both the DFS and IS groups but to a lesser degree in the DFS group (13). In a field trial of another DFS in Indian tea pickers, hemoglobin concentrations and work output improved (32). However, the authors provided no details on the iodine and iron compounds in the DFS and no data on color or iodine stability. The Micronutrient Initiative (14, 31) developed a DFS containing potassium iodide coated with maltodextrin and ferrous fumarate. Iron absorption from this DFS in iron-enhancing and iron-inhibiting meals was 13.5% and 4%, respectively (12). However, nonencapsulated ferrous fumarate added to low-grade salt in developing countries causes unacceptable dark brown color changes (15).

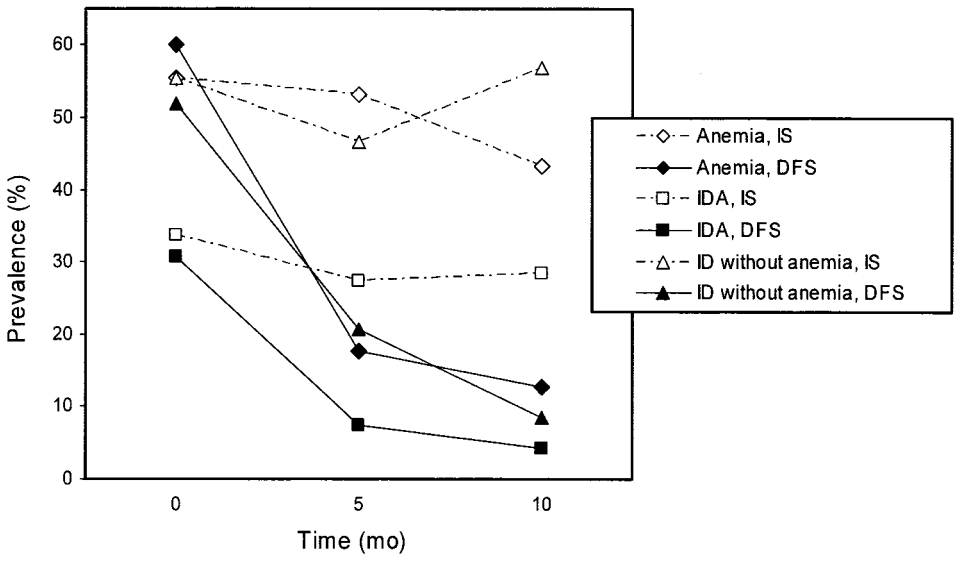

FIGURE 1. Prevalence of anemia, iron deficiency anemia (IDA), and iron deficiency (ID) without anemia in 6–15-y-old children receiving dual-fortified salt (DFS) containing iron and iodine or iodized salt (IS) for 10 mo. By logistic regression, in comparison with the children who received the IS, those who received the DFS had significantly decreased prevalences of anemia, IDA, and ID without anemia. The difference between the groups increased with time ($P < 0.001$; comparison of time-and-treatment model with time-only model).

Table 5
Urinary iodine concentrations, thyroid volumes, and goiter prevalence in the iodized salt (IS) and dual-fortified salt (DFS) groups over 10 mo

	Urinary iodine (μg/L)[1]		Thyroid volume[2]		Goiter prevalence[3]	
Time (mo)	IS group	DFS group	IS group	DFS group	IS group	DFS group
	μg/L		*mL*		*n (%)*	
0	12 (2–70)[4]	10 (3–121)	8.3 ± 3.4[5]	8.5 ± 3.7	59[6] (70)	54 (72)
5	74 (2–239)	71 (4–273)	7.7 ± 3.1 (−7.2 ± 10.8)	7.2 ± 3.0[6,7] (−15.4 ± 9.4[6,7])	49 (59)	41 (54)
10	104 (22–1784)	97 (17–1356)	6.9 ± 2.2 (−16.3 ± 7.4)	5.9 ± 2.3[8,9] (−29.6 ± 8.6[8,9])	42 (51)	29 (39)

[1] Significant main effect of time, $P < 0.001$.
[2] Percentage change from 0 mo in parentheses. Significant treatment × time interaction, $P < 0.001$ (ANOVA).
[3] By logistic regression, the group difference increased with time ($P < 0.01$; comparison of time-and-treatment model with time-only model).
[4] Median; range in parentheses (all such values).
[5] $\bar{x} \pm$ SD (all such values).
[6,8] Significantly different from baseline: [6]$P < 0.05$, [8]$P < 0.01$.
[7,9] Significantly different from the IS group: [7]$P < 0.05$, [9]$P < 0.01$.

We recently showed the efficacy of a DFS containing ferrous sulfate encapsulated with partially hydrogenated soybean oil (4). In a 40-wk trial in schoolchildren, mean hemoglobin concentrations increased by 14 g/L, and the prevalence of IDA decreased from 35% to 8%. In this previous efficacy study, although the iodine was stable in the DFS, iron encapsulation did not prevent color changes; the DFS developed a yellow color when the moisture content of the salt was 3–4% (4). We recently tested the stability of 16 different forms of encapsulated iron in local salt in West and North Africa (15) and found that encapsulated ferrous iron caused unacceptable color changes in salt. Current encapsulation technology, which uses hydrogenated plant oils, does not sufficiently reduce moisture penetration and iron solubility, and capsule integrity is further compromised by mechanical abrasion during salt mixing. An additional barrier to the use of encapsulated iron in salt fortification is the relatively low melting point of the capsules (45–65 °C), which may cause unwanted sensory changes during food preparation (33). Finally, high processing costs for encapsulation may limit applications in developing countries.

FePP has a white color and produces negligible color change when added to low-grade, high-moisture salt in West and North Africa, even during prolonged storage (15). Ferric iron is relatively stable and nonreactive in the presence of potassium iodate, which is the iodine compound recommended for salt fortification in developing countries (34). When FePP is added to salt containing potassium iodate, iodine stability is comparable with that in IS (15). In contrast, ferrous iron reacts strongly with potassium iodate to form ferric oxides and iodine (I_2), which causes color change and iodine losses. For these reasons, FePP has clear advantages as a component of a DFS.

However, FePP is a poorly soluble iron compound, and its low bioavailability—only 30–50% of ferrous sulfate—reduces its nutritional value (8, 9). Stable-isotope studies have reported that the RBV of FePP with MPSs > 10 μm is only one-third that of ferrous sulfate and ferrous fumarate (35, 36). Particle size is an important determinant of iron absorption from poorly soluble iron compounds in foods. Decreasing the particle size of elemental iron powders by 50–60%, to an MPS of 7–10 μm, increases iron absorption by ≈50% in rats (37, 38). In a human study, iron absorption from hydrogen-reduced elemental iron with particle sizes between 5 and 10 μm was comparable with that of iron sulfate (39). Similarly, reducing the particle size of FePP increases its absorption. Conventional grinding can decrease the MPS of FePP to 2–3 μm. Further reduction in MPS to <1 μm is possible by generating FePP particles in aqueous solutions and adding emulsifiers to prevent agglomeration (40). In a human study, decreasing the MPS of FePP from 8.5 to 6.7 μm increased its RBV compared with iron sulfate from 36% to 52% (36). In a stable-isotope study, the RBV of a dispersible FePP with an MPS of ≈0.5 μm was comparable with that of ferrous sulfate (16). Recent hemoglobin repletion studies in rats have shown that the RBV of FePP with MPSs of 2.5 and 0.5 μm is ≈70% and 95% that of ferrous sulfate, respectively (R Wegmüller, personal communication, 2004). Although FePP with an MPS of 0.5 μm may be more bioavailable, we chose to use FePP with an MPS of 2.5 μm in this study because it is less expensive and has better handling characteristics and because the commercially available FePP with an MPS of 0.5 μm is a patented compound (40). In the single previously reported efficacy study of FePP (41), Pakistani infants received a wheat- and milk-based food supplement fortified with either FePP, ferrous fumarate, or control from 4 to 12 mo of age. Both iron-fortified supplements contained 7.5 mg Fe/100 g; the total daily iron dose was ≈3 mg. After 12 mo, the iron status of the 2 iron groups was not significantly different, and the iron status of both groups was higher than that of the control group (mean hemoglobin: 10.4 compared with 9.9 g/dL; mean SF: 13.3 compared with 8.8 μg/L; $P < 0.05$).

In the present study, micronized FePP with an MPS of 2.5 μm showed good bioavailability. The total dose of iron delivered by the DFS during the 10-mo trial was ≈5.4 g, on the basis of a study period of 300 d, mean salt intakes of 9 g/d in children, and a fortification concentration of 2 mg Fe/g salt. The mean increase in total body iron in the DFS group was ≈106 mg, whereas the mean decrease in body iron in the IS group was ≈10 mg (26). On the basis of these data, the mean daily gain in body iron in the DFS group was ≈0.38 mg, which suggests that ≈2% of the fortification iron was absorbed over the 10-mo trial.

Several factors favorably contributed to iron efficacy in this study. We provided a relatively large (≈18 mg/d) iron dose to growing children with poor iron status. The children ate 3 main meals, as well as mid-morning and mid-afternoon snacks, 7 d/wk, and all meals and snacks contained appreciable amounts of salt. Thus, absorption was probably enhanced by iron delivery in

repeated small doses throughout the day, because fractional absorption of nonheme iron increases with decreasing dose (42). In this region, IDA is primarily due to low dietary iron bioavailability and not increased iron losses. There is no malaria in northern Morocco, and hookworm and other intestinal parasites that cause blood loss are rare (M Bousfiha, personal communication, 2002). Moreover, the low prevalence of infection or inflammation in the children (only ≈5% had elevated CRP) sharpened our ability to rely on SF and other iron indexes to clearly define IDA and detect changes in iron status (25, 43).

The DFS was also an effective vehicle for iodine. The iodine concentration in the DFS was not significantly different from that in the IS over 6 mo of storage. At baseline, the children were severely iodine deficient: the median UI concentration was 12 μg/L, and the goiter rate was 70%. The iodine in both salts was highly bioavailable: median UI concentrations increased by 10 mo in both groups to values indicative of iodine sufficiency. Thyroid volume and goiter prevalence decreased significantly in both groups, but consistent with previous reports, the addition of iron to IS improved iodine efficacy (5–7). Compared with the IS group, the DFS group had a significantly greater reduction in thyroid size and goiter prevalence. After 10 mo of salt use, which included the damp winter season, 98–100% of households receiving the DFS rated its color and taste as acceptable. The DFS did not change color during 6 mo of storage, and there was only a small absolute color difference (ΔE value of ≈7) compared with the IS. On the basis of studies in West and North Africa, the color of a DFS is usually acceptable if the absolute color difference (ΔE value) compared with the local IS is <10 (15).

In previous iron-fortification trials that clearly improved iron status in target populations (44–48), the iron-fortified food was consumed with an enhancer of iron absorption (ascorbic acid or EDTA), which was added to overcome absorption inhibitors. In the present study, despite the high phytic acid content of the diet, iron fortification of salt without an enhancer significantly improved iron status. Our findings indicate that micronized FePP as part of DFS is an effective iron-fortification strategy and may provide new opportunities for the global control of IDA. The cost of the micronized FePP used in this study is ≈4 times the cost of ferrous sulfate on a per iron basis (W Vogl, personal communication, 2004). Because the performance of DFS may vary depending on climate, salt quality, and dietary habits, we are currently repeating these studies in West Africa, where environmental conditions are harsher and the salt quality is poorer.

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Each of the authors contributed to the study design. The fieldwork and data collection were performed by MBZ, RW, FR, CZ, NC, and MS. MBZ, RW, FR, CZ, TT, and RFH completed the final laboratory and data analysis. The statistical analysis was done by MBZ. The first draft of the manuscript was written by MBZ, with editing by all the remaining authors. None of the authors had any financial or personal conflicts of interest in regard to this study.

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