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Optimization of Unit Operations for Microencapsulating Ferrous Fumarate During Scale-Up of Double Fortification of Salt with Iron and Iodine

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Abstract

Objectives: This study evaluates factors responsible for the floating of iron premix in double fortified salt (DFS), which initially affected the large-scale implementation of the salt fortification program in India, and provides solutions to the scale-up of the technology.
Materials and Methods: To mitigate this time-sensitive scale-up challenge. First, the iron premix samples were obtained from the industrial scale-up pilot studies in India, evaluated for the impact of the amount of coating material (5 per cent, 7.5 per cent, and 10 per cent (in weight)), type of formulation (soy stearin, SEPIFILM and hydroxypropyl methylcellulose), amount of titanium dioxide (25-35 per cent (in weight)) used for color masking; Second, we studied the effect of change in the composition of the coating, from 10 per cent (in weight) soy stearin to a double coat with 5 per cent (in weight) hydroxypropyl methylcellulose and 5 per cent soy stearin or 10 per cent soy stearin and 1 per cent (in weight) lecithin mixture, on particle density, floating or sinking property of the iron premix, and on the stability of iodine in the DFS.
Results: It was observed that the hydrophobic nature and the amount of soy stearin used for coating caused the floating issue. The double coating with 5 per cent hydroxypropyl methylcellulose and 5 per cent soy stearin was preferred because lecithin in soy stearin enhanced the moisture-aided adverse interaction between iron and iodine. Shelf-life storage studies proved over 80 per cent iodine retention after 12 months of storage in the DFS formulated with iron premix double-coated with hydroxypropyl methylcellulose and soy stearin.
Conclusion: This proffered solution enabled the full implementation of the double fortification program in India.

Keywords: Salt fortification; spray coating; soy stearin; hydroxypropyl methylcellulose; iron deficiency.

Introduction

Salt iodization remains one of the most successful and vital contributions in the field of food fortification. Its effect on reducing the global prevalence of iodine deficiency is unprecedented; salt has been viewed as one of the most effective vehicles for delivering iodine and has the potentials to deliver multiple micronutrients (Allen *et al.*, 2006). Hence, many researchers have formulated salt-containing iron and iodine (double fortified salt, or DFS) to simultaneously combat the prevalence of iron and iodine deficiencies (Rao, 1994; Zimmermann *et al.*, 2004; Li *et al.*, 2009). While the sources of iodine fortificants are limited to calcium iodate, potassium iodate, and potassium iodide, iron fortificants are numerous. The taste and bioaccessibility considerations in these fortificants made by the Food Engineering Research Group at the University of Toronto choose ferrous fumarate as iron fortificant (Diosady *et al.*, 2002). However, the ferrous fumarate color and its potential interaction with iodine in the fortified salt necessitates color masking and microencapsulating ferrous fumarate (iron premix) before mixing it with iodized salt.

The process developed by the Food Engineering Research Group at the University of Toronto for making the iron premix involves the agglomeration of ferrous fumarate, size screening, color masking, and coating. The first generation of the technology was a one-step approach to all these processes (agglomeration, color masking, and coating) using a pan coater, fluidized bed, or spray dryer. The research and development of the technology spans over 20 years with multiple generations of product improvement and cost reduction. Unfortunately, the first-generation technology developed was not further pursued for several reasons. The pan coater and fluidized bed require a lot of soy stearin; this caused the floating of coated iron particles in water. Also, the particles were irregular in shape. Finally, using a spray dryer limited the iron loading percentage in the premix; it was challenging to mask the spray-dried particle white enough to match the salt color and effectively coat the iron particles.

In the second generation of the technology, the four initial unit operations were split, and there was an expansion of the size screening operation, which now included a cutting step. Several efforts have been reported to optimize the unit operations to make the iron premix for the salt's double fortification (Li *et al.*, 2011; Yadava *et al.*, 2012). These studies proposed the use of durum semolina as a binder and vegetable shortening as a lubricant for extrusion, titanium dioxide as a color masking agent, and hydroxypropyl methylcellulose (HPMC) as the surface over-coating material to replace the hydrophobic coating (soy stearin) in the original formulation (the first-generation technology). The color masking of the brown iron extrudate is achieved by dusting–rubbing–firming the TiO_2 on the extrudate's surface. Then, the TiO_2 loosely held by a weak electrostatic force is held firmly on the extrudate surface by applying an HPMC coat using a fluidized bed spray. This coat also ensures the iron premix's physical integrity. In addition, it prevents the chemical interaction between iron and iodine when the iron premix is blended in iodized salt to make DFS. The optimization involved careful considerations for selecting materials and the amount used to achieve the iron's desirable bioaccessibility in the premix; it is needless to have a perfectly coated iron premix whose iron is not bioavailable to perform its metabolic functions. Thus, the coat was designed to release the encapsulated iron after the stomach digestion phase by exposing it to pH 1.

The optimizations of the cold-forming extrusion-based microencapsulation of ferrous fumarate by Li *et al.* (2011) and Yadava *et al.* (2012) led to an established laboratory-scale innovation to make stable and bioavailable iron premix for the double fortification of salt. The technology is easily adaptable to the traditional process of salt iodization. The making of iron premix is entirely separate from the traditional method of making iodized salt. Hence, that is the part of the technology that was scaled up by several pilot testing trials that eventually achieved full commercialization in India. During the pilot trials, the HPMC coat designed in the laboratory was changed to a soy stearin coat because of the pilot plant region's high humidity. This change makes the premix surface hydrophobic, preventing adverse moisture-aided interaction between iron and iodine in the DFS, leading to iodine loss.

However, the iron premix coated with soy stearin floated in the water and other liquid foods during the technology's initial scale-up trials. This problem led to the iron premix being washed away during food preparation. This study aims to understand the factors responsible and proffer solutions to the problems. First, we evaluated the optimal amount of coating material that can effectively prevent the interactions between iodine and iron in the DFS produced at the pilot plant. Next, we evaluated the particle and bulk density of the premixes coated with several materials to elucidate the factor responsible for the premix's floating while proffering solutions to the problem. This study is crucial for successfully implementing the DFS to combat iron and iodine deficiencies in large populations in India. This technology is currently reaching over 60 million people in India and can be expanded to other countries affected by micronutrient deficiency (Diosady *et al.*, 2018, 2019).

Materials and Methods

Material

Ferrous fumarate, soy stearin, HPMC, durum semolina, Crisco Shortening, and titanium (IV) oxide, used in the production of iron premix in the Food Engineering Laboratory at the University of Toronto, were obtained from Dr. Paul-Lohmann Chem (Emmerthal, Germany), JVS Food Pvt. Ltd. (an industrial partner for iron premix production in Jaipur, India), Dow Chemical Company (Midland, MI, USA), Unico Inc. (Concord, Ontario, Canada), J.M. Smucker Co. (Orrville, OH, USA), and ACROS Organics (Fairlawn, NJ, USA), respectively. The iron premix was received from the pilot scale plant in India (JVS Food Pvt. Ltd.) after completing the first pilot study in June 2015. The formulation design leading to these final premixes was reported separately (unpublished). While the final premixes (batches 1–9) were used for the stability study in this research, the uncoated iron extrudates (dark brown core) obtained from JVS Food Pvt. Ltd. from the same pilot trial were used for laboratory studies of whitening and coating (material levels and applying methods) in the second part of this research. Potassium iodate (used for laboratory preparation of iodized salt for shelf stability study) was obtained from Sigma-Aldrich Chem (Oakville, Ontario, Canada). Potassium iodide, used for the analysis of iodine, was obtained from Caledon Lab Chem (Georgetown, Ontario, Canada). Sulfuric acid and starch indicator, used for iodine analysis, were obtained from EMD (Oakville, Ontario, Canada) and Caledon Lab Chem (Georgetown, Ontario, Canada). All chemicals used for the fortification of salt were food-grade. In contrast, those used for analysis were American Chemical Society grades. Refined salt (approximately 400 µm diameter) was obtained from Sifto Canada Corp. (Mississauga, Ontario, Canada).

Formulation of iron premix

The technology described by Li *et al.* (2011) and Yadava *et al.* (2012) was scaled up at JVS Food Pvt. Ltd. The ferrous fumarate ratio to durum semolina was kept as recommended (80:20) for all batches except for batch 1 at 75:25; TiO_2 for color masking was 25–35 per cent (in weight); the type of coating material and amount used was also varied. In batches 1–3, the performances of three different coating materials were compared. The materials were hydrophilic (HPMC), hydrophobic (soy stearin), and Sepifilm, a ready-to-use gastro-soluble film-coating agent primarily designed for pharmaceutical applications with a high cost. In batches 4–6 and 7–9 (two comparable groups), HPMC was used for coating with a repeated increase from 5 per cent to 7.5 per cent and then 10 per cent (in weight), whereas the color-masking of TiO_2 was set at 35 per cent and 30 per cent, respectively. The two groups of batches 4–6 and batches 7–9 were designed to evaluate the combined effect between the amounts of TiO_2 and hydrophilic coating material (HPMC) that can effectively prevent interaction between iron and iodine in the fortified salt, targeting an optimal usage level of coating materials thus lower costs.

The premix coat designs' effectiveness to prevent interaction between iron and iodine in the DFS was evaluated. A solution of 3.37 g/L potassium iodate (2.5 mL) is sprayed onto 1 kg salt, leading to 5×10^{-5} iodine in salt. The iodized salt was dried overnight. The premix samples were mixed with the iodized salt at a ratio of 1:200. This ratio corresponded to having approximately 1×10^{-3} and 5×10^{-5} of iron and iodine, respectively, in salt. The percentage retention of iodine was evaluated after 2 months and 6 months of storage. The salt samples were stored at 25 °C and 45 °C, 60–70 per cent relative humidity (RH).

Effect of coating materials on the floating/sinking property of iron premix

The iron extrudate (dark brown core) obtained from JVS Food Pvt. Ltd., India was color masked with 25 per cent TiO_2 and coated with 10 per cent (in weight) soy stearin or HPMC. The particle and bulk densities of the iron premixes were determined. The impact of changing the coating materials on the floating property of the iron premixes was evaluated. Because the floating problem was not observed in the initial iron premix coated with only 5 per cent soy stearin and given the emulsifying property of lecithin (plant-based), two approaches were attempted in the laboratory to understand and solve the floating problem of iron premix: coating with a mixture of lecithin and soy stearin, and dual coating with 5 per cent HPMC and 5 per cent soy stearin. In total, for this batch, four premix samples were made in the laboratory. Also, a premix was obtained from the pilot plant in India; the premix was double coated with 5 per cent HPMC and 5 per cent soy stearin.

The soy stearin coating formulation was melted in a thin-layer chromatography sprayer flask; dichloromethane was added to make 80 g/L soy stearin. For HPMC, warm water was used to wet its surface before it was dissolved in a 50:50 ethanol–dichloromethane solvent system to make 25 g/L HPMC. Other proportions of the ethanol–dichloromethane solvent system were also evaluated. A heated hair dryer was attached to the pan coater base to aid the evaporation of the solvent. The coating solution was applied at about 3 mL/min with the thin-layer chromatography sprayer.

DFS was formulated with the five premix samples, as previously described. The stability of iodine in the salt was evaluated for 2, 6, and 12 months. After 2 months, the evaluation of iodine's stability in the DFS formulated with premix-coated lecithin and soy stearin was stopped due to the drastic loss of iodine in the salt. The salt samples were stored at 25 °C and 45 °C, 60–70 per cent RH.

Determination of the total iron and coating integrity of premix samples

The iron premix was digested with concentrated nitric acid in a microwave digester (John Morris Scientific Pty Ltd., Balwyn, Victoria, Australia), as described by Modupe and Diosady (2021). The solution was reconstituted with reverse osmosis (RO) water and filtered. The amount of iron exposed on the premix samples' surface was used as a yardstick for judging the coating's integrity. Ethylenediaminetetraacetic acid (EDTA) is an iron chelator; the 50 g/L EDTA was used to dissolve iron on the premix's surface for 5 minutes before the solution penetrates the premix's iron core. The solution was filtered.

The amount of iron in the filtrates was determined using inductively coupled plasma - optical emission spectrometry. The amount of iron on the surface of the premix is expressed as a percentage of the total iron in the premix.

Determination of the densities

An empty 25 mL scintillation vial was weighed (W_1). Next, it was filled with the premix samples and was tapped until no apparent volume change was observed. The weight of the sample-filled vial was then recorded (W_2). Then, the vial was emptied and filled with water and weighed (W_3). The bulk density of the sample was then calculated as follows according to Equation (1):

$$\rho_B = \frac{W_2 - W_1}{W_3 - W_1} \times \rho_W \tag{1}$$

where ρ_B is the bulk density in g·cm^{-3} and ρ_W is the water density in g·cm^{-3}.

After bulk density was determined as described using Equation (1), hexane was added dropwise to determine the void volume in the sample-filled flask. The weight of the flask was measured (W_4), and Equation (2) was used to calculate the particle density:

$$\rho_P = \frac{(W_2 - W_1)}{\left(\frac{W_3 - W_1}{\rho_W}\right) - \left(\frac{W_4 - W_2}{\rho_H}\right)} \tag{2}$$

where ρ_P is the particle density in g·cm^{-3}; ρ_W is the water density equaling 1 g·cm^{-3}; and ρ_H is the hexane density equaling 0.66 g·cm^{-3}.

Surface morphology of premix samples

The iron premix samples' surface morphology was determined by scanning electron microscopy (SU-3500 VP SEM, Hitachi High-Technologies Corp., Tokyo, Japan), as described by Singh *et al.* (2018).

Iodine analysis

The concentration of iodine in the salt samples was determined by iodometric titration as described by the Association of Official Analytical Chemists, Arlington, VA, USA (Williams, 1984).

Statistical analysis

All chemical analyses for each sample were conducted in triplicate, and the results were calculated and expressed as a means±standard deviations for each of the measurements. The data were subjected to one-way analysis of variance (ANOVA) using IBM SPSS software (IBM Corp., Armonk, NY, USA), and the differences among the means were considered significant at $P<0.05$.

Results and Discussion

The cold-forming extrusion-based microencapsulation of ferrous fumarate has four primary operations—particle agglomeration, cutting and size matching, color masking, and coating. The technology ensures four main functions in DFS: it prevents chemical interaction between iron and iodine, masks the brown color of ferrous fumarate, prevents segregation in salt by matching the size of the iron premix to the size of salt, and ensures the encapsulation can withstand pH 1 (simulated stomach pH). Overall, it achieves a uniform and visually indistinguishable dispersion of 0.5 per cent (in weight) iron premix in iodized salt. Our attempts to address the challenges during scale-up of the iron premix particle agglomeration for size matching were solved by mechanically adjusting the extruder and a spheronizer used for cutting and shaping the extrudate into 300–600 µm spheres. The systematic study (of the coating challenges and translation from laboratory to scale-up) forms the basis for this study.

Effect of coating composition on the total iron content and the amount of exposed iron on the surface of the premix (from pilot plant)

Based on the work carried out by Li *et al.* (2011) and Yadava (2012) at the University of Toronto, 25–35 per cent TiO_2 was used for color masking, and 5–10 per cent of coating materials were used for coating at the pilot plant. The total iron in the premix samples of the nine batches was obtained from the pilot plant and analyzed; the result was used to calculate iron's percentage composition in the premix. There was very little difference in the iron content of the nine premix samples. The premix contained 18–20 per cent (in weight) iron (Figure 1). The result is consistent with previous studies (Li *et al.*, 2011; Yadava *et al.*, 2012) and the premix formulation's material balance estimate. Given the percentage of iron in the premix, approximately 1 iron in the fortified salt is achievable by adding 5 g of iron premix to 1 kg of salt.

The premix coating performs two functions—it holds the whitening agent in place and prevents the chemical interaction between iron and iodine, leading to the iodine's loss in the fortified salt(Yadava *et al.*, 2012). Therefore, an ideal coating must result in a premix with little or no iron exposed on its surface. The amount of iron exposed on the surface of the premix samples can determine the coating's integrity. Less than 5 per cent of the iron in the premix was exposed on the premix's surface (Figure 1). This value is close to those (<10 per cent) reported by Yadava *et al.* (2012) and implies a good coating. The amount of iron on the premix's surface correlates with the iodine lost from DFS after two months of storage (Figure 2A). Comparatively, coating with 5 per cent soy stearin (fat) was better than coating with 5 per cent HMPC for retaining iodine in salt (Figure 1, comparing premix sample 3 vs. premix sample 7). The amount of iron exposed on the surface of the premix samples (comparing premix sample 4 vs. sample 7; premix sample 5 vs. premix sample 8; and premix sample 6 vs. premix sample 9 in Figure 1) showed that increasing the amount of TiO_2 (30 per cent to 35 per cent) used for color masking may negatively affect the effective coating of the premix with HPMC. The use of 10 per cent HPMC (premix sample 9 in Figure 1) resulted in less iron on the premix's surface when 30 per cent TiO_2 was used for color masking. Hence, 10 per cent coating material and color masking with 30 per cent TiO_2 were subsequently adopted. From the result, an excessive amount of TiO_2 (more than 30 per cent (in weight)) as a color masking agent may not be necessary. It may lead to a larger particle size requiring more coating material for effective coating. More so, it may increase the cost of production. The pilot scale-up study was conducted between 2016 and 2019, and TiO_2 was a Generally Recognized as Safe food ingredient. Considering the recent European Food Safety Authority report in May 2021 with concerns about using TiO_2, the University of Toronto salt fortification team has started exploring alternative whitening agents to replace TiO_2 for future improvements of the iron premix coating.

Effect of coating composition on the stability of iodine in double fortified salt

The adverse interaction between iron and iodine in DFS that leads to the loss of iodine was minimized by coating the ferrous fumarate extrudate. Without coating, the chemical interactions could lead to the iodine displacement from the iodate, which is then lost by sublimation. The impact of the coating composition of

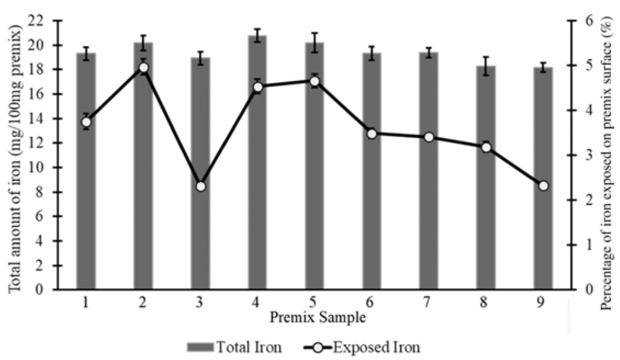

Figure 1. Effect of the coating composition on the iron content and iron exposed on the surface of iron premix samples 1–9 varied in the amount of TiO_2 and coating materials used. Samples 2–3 and 7–9 were color-masked with 30 per cent TiO_2, while samples 1 and 4–6 were color-masked with 25 per cent and 35 per cent TiO_2, respectively. Sample 1 was coated with 10 per cent HPMC; sample 2 was coated with 10 per cent Sepifilm; sample 3 was coated with 5 per cent soy stearin; samples 4 and 7, 5 and 8, and 6 and 9 were coated with 5 per cent, 7.5 per cent, and 10 per cent HPMC, respectively. Values are average of four replicates±standard deviation.

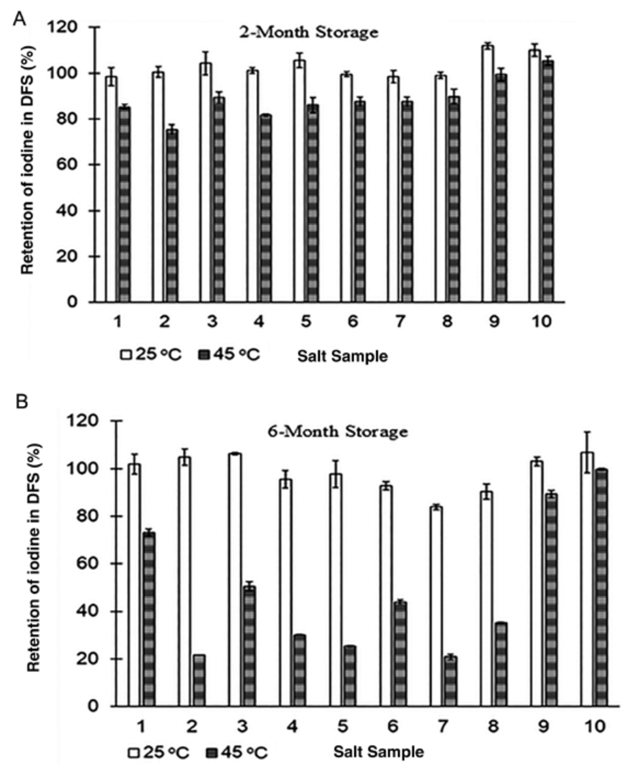

Figure 2. The retention of iodine in DFS at 2 months (A) and 6 months (B) of storage. Salt samples 1–9 were DFS samples made with corresponding premix samples 1–9, while sample 10 is iodized salt. Values are average of four replicates±standard deviation. DFS, double fortified salt.

the premix obtained from JVS Foods Pvt. Ltd. on iodine stability in DFS was investigated. In all the salts stored for 2 months, at least 75 per cent iodine was retained irrespective of storage conditions (Figure 2A). The iodine retention in DFS at the sixth month of storage at 45 °C (Figure 2B) clearly showed that 10 per cent coating was adequate if only 30 per cent of TiO_2 is used for color masking (salt sample 6 vs. salt sample 9 in Figure 2B). The iodine retention in DFS formulated with iron premix color masked with 30 per cent TiO_2 and coated with 10 per cent HPMC (salt sample 9) was comparable to the iodine retained in the iodized salt (salt sample 10). Hence, the extrudate color masked with 30 per cent TiO_2 and coated with 10 per cent HPMC was the best formulation in this test series.

The results were consistent with the coating integrity observations, which showed that iron premix coated with soy stearin (sample 3) and 10 per cent HPMC (sample 9) had the least exposed iron. Hence, it is not surprising that the DFS formulated with the premix sample 9 were among the three salts with the highest iodine retention after 6-month storage at 45 °C.

Four factors impacted the loss of iodine from the salt—the amount of TiO_2 used to mask the brown color of the iron extrudate, the properties of the material used to coat the premix, the amount of coat, and storage temperature. The use of more than 30 per cent TiO_2 (as seen with salt sample 6 vs. salt sample 9 in Figure 2B) did not result in effective coating with 10 per cent of the coating material. Excess TiO_2 may require more coating material to hold the TiO_2 in place on the premix's surface and form an effective physical barrier between iron and iodine in a DFS. The use of 10 per cent coating material resulted in a better physical barrier between iron and iodine than when 5 per cent or 7.5 per cent was used. Soy stearin, being hydrophobic, was a better coat than HPMC. When the same amount of TiO_2 and coating material is used (30 per cent and 5 per cent, respectively), soy stearin prevented the interaction between iron and iodine better than HPMC (as seen with salt sample 3 vs. salt sample 7). Its hydrophobicity impedes moisture penetration to the iron core. The increase in the storage temperature of the salt accelerated the impact of these factors.

Formulation of coating solutions

Pan coater was used to coat the color-masked extrudate to simulate the drum coater used at the pilot scale. The coating solution must be very volatile for it to be applicable for the pan coating. Dichloromethane was sufficiently volatile for the pan coating. While melted soy stearin is soluble in dichloromethane, HPMC is not. HPMC wetted with water is soluble in absolute ethanol, which is not volatile enough for the pan coating operation. Hence, an ethanol–dichloromethane solvent system was used. First, the miscibility of the two solvents was evaluated by mixing different proportions of the solvents—the more ethanol, the better the miscibility. Ultimately, a ratio of 1:1 of the two solvents was used to make a solution of HMPC. A higher proportion of dichloromethane resulted in the HPMC precipitation out of the solution (Figure 3). The remarks on the use of other proportions of the solvent system are presented in Table 1.

Impact of coating material on physical properties of the premix

At the pilot scale plant, 30 per cent TiO_2 was used for color masking, and 10–15 per cent soy stearin was used as the coating material, given that soy stearin was comparatively better than HPMC as an effective coat (sample 3 vs. sample 7 in Figure 3B), that 10 per cent coating material is the optimal amount, and that using more than

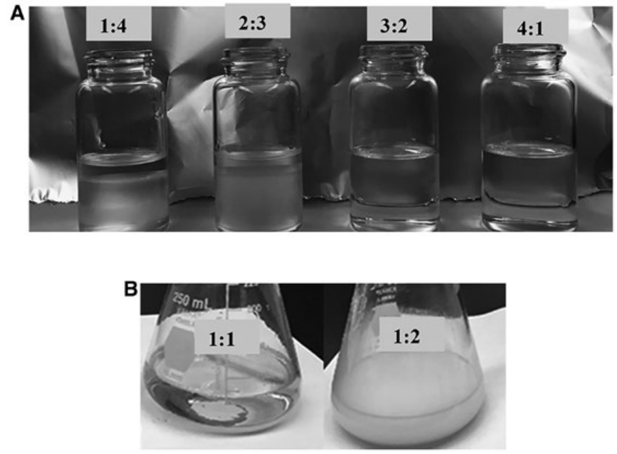

Figure 3. The impact of the proportions of ethanol to dichloromethane on the miscibility of the solvents (A) and the solubility of HMPC in ethanol–dichloromethane solvent (B).

Table 1. Overall remarks on the ratio of solvents used

Dichloromethane (%)	Ethanol (%)	Remarks
20	80	• Dissolved HPMC • Wetted the premix • Premix clumped
40	60	• Dissolved HPMC • Wets the premix • Premix clumped
50	50	• Dissolved HPMC • Did not wet the premix • Premix did not clump • Formed a uniform suspension of folic acid
60	40	• Slightly dissolved HPMC • Folic acid precipitated
80	20	• Did not dissolved HPMC

HMPC, hydroxypropyl methylcellulose.

Table 2. Impact of coating material on the particle and bulk density and floating properties of the iron premix

Sample	Particle density ($g \cdot cm^{-3}$)	Bulk density ($g \cdot cm^{-3}$)	Floating
10% SS	2.09±0.10	1.08±0.06	Significant
10% HPMC	2.19±0.21	1.06±0.07	Insignificant
10% SS+Lecithin	2.11±0.20	1.21±0.15	Insignificant
5% HPMC, 5% SS	2.06±0.12	1.11±0.02	Insignificant

HMPC, hydroxypropyl methylcellulose; SS, soy stearin.

30 per cent TiO_2 does not support effective coating. However, the premix made with this formulation floats in water (observation was from a field test of the DFS in India). Two factors may be responsible for this effect: the density and the hydrophobicity of soy stearin.

We determined the densities of the premix coated with 10 per cent soy stearin that floats and premix coated with 10 per cent HPMC that does not float in water. There was no significant difference in the bulk and particle densities of premix coated with 10 per cent HPMC and those coated with 10 per cent soy stearin made in the laboratory (Table 2). Hence, surface tension due to

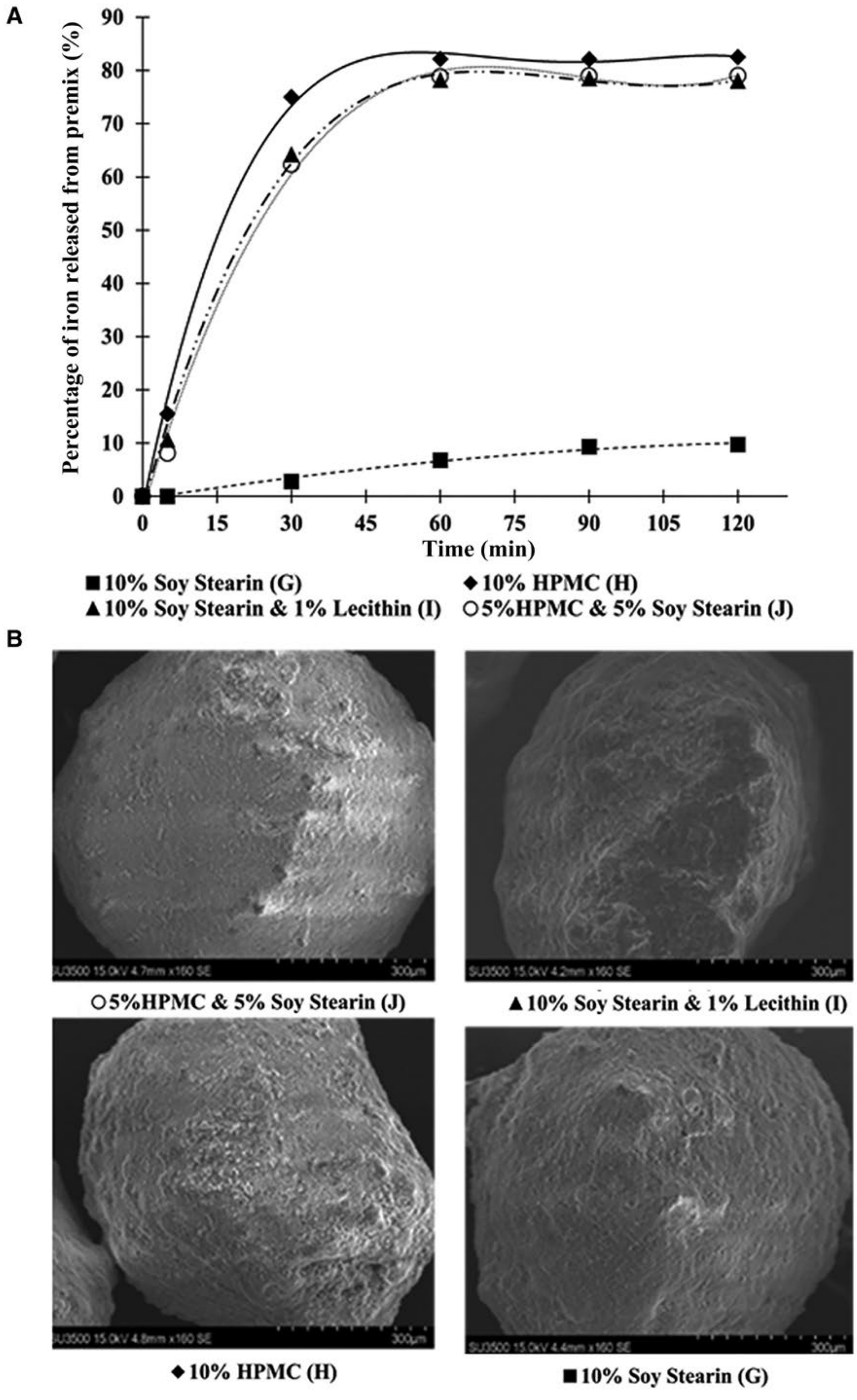

Figure 4. Time profile of iron release from the premix at pH 1 (A) and microscopic imaging of the premix (B). HPMC, hydroxypropyl methylcellulose.

soy stearin's hydrophobicity but not density was responsible for the floating observed. The result is not surprising as the amount of TiO_2, which has the highest density of all materials used to formulate premix, is the same in all these premix samples.

We evaluated two approaches for solving the floating problem: dual coating with 5 per cent HPMC and 5 per cent soy stearin, and coating with an 11 per cent mixture (10:1) of soy stearin and lecithin. The two approaches significantly improved the sinking property of the premix

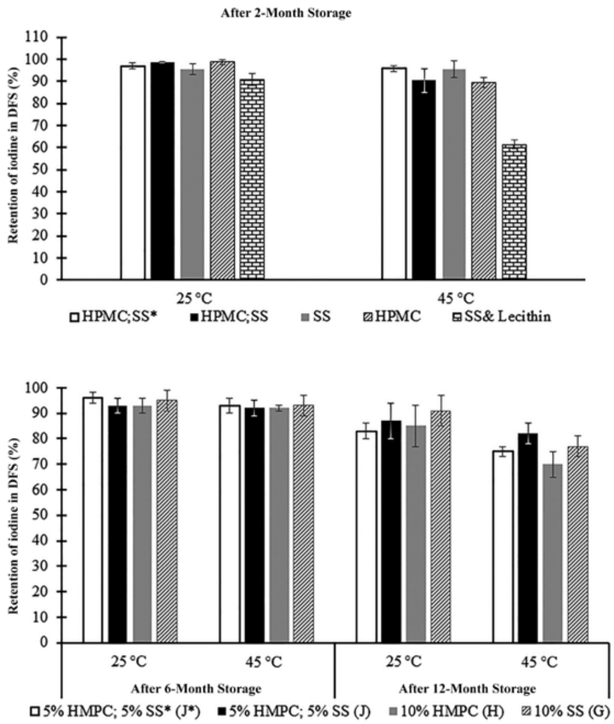

Figure 5. Stability of iodine in DFS after 2, 6, and 12 months storage. *Sample obtained from pilot scale. DFS, double fortified salt; HPMC, hydroxypropyl methylcellulose; SS, soy stearin.

(Table 2). The ability of added lecithin, an emulsifier, to solve the sinking problem is also an indicator of the hydrophobic nature of soy stearin as the factor responsible for the initial floating of premix. The sinking observed with premix coated with just 5 per cent soy stearin suggests that the soy stearin's bulkiness in the premix may be a factor. The lecithin added to soy stearin seems to impact its hydrophobicity; hence, it adversely affected iodine's stability in the DFS after storage for 2 months. More so, we adopted the coating with 5 per cent HPMC and then 5 per cent soy stearin because the soy stearin can still provide significant resistance to moisture. While this increased the premix formulation by one step at the laboratory scale, this does not significantly impact the industrial scale. Both HPMC and soy stearin coats can be applied on iron extrudate with a drum coater. Thus, the 5 per cent HPMC and 5 per cent soy stearin formulation was easily scaled up from laboratory- to industrial-scale production.

There were no significant differences in the four premix samples' surface morphology from the scanning electron microscopic imaging (Figure 4B). This result and the corroborated observations from the iodine stability test may indicate a perfectly coated premix. In addition, there were no color changes in food samples prepared using DFS salt—by independent consumer acceptability and sensory studies conducted by the University of Delhi (Diosady *et al.*, 2018).

Although the premix was designed to disintegrate with many cooking techniques, there are a few cases where salt may be used without cooking or a few cooking techniques that may not disintegrate the premix. Hence, iron bioaccessibility, a vital parameter to access the release of iron from the premix, was evaluated. Only 10 per cent iron was released from the premix coated with 10 per cent soy stearin after 2 hours of dissolution in hydrochloric acid solution (pH 1) compared to over 75 per cent iron released from other premix coated with other formulations (Figure 4A). Hence, aside from floating problems caused by coating with 10 per cent soy stearin, it may also prevent iron release from the premix when it is not disintegrated by the cooking method before ingestion. It should be noted that the pH of the stomach may fluctuate; however, the combined action of the stomach acidity and enzyme action should outperform the stimulated iron release.

Effect of change in coating material on the stability of iodine in double fortified salt

Because the premix sample coated with soy stearin and lecithin caused a drastic loss of iodine in the DFS after 2 months of evaluation, it was not used further. In addition to the other three premix samples prepared in the laboratory, industrial premix was obtained from the pilot scale (coated with 5 per cent HPMC and 5 per cent soy stearin, to compare a similar premix prepared in the laboratory) to formulate DFS. The result showed that 10 per cent coating material provided an effective barrier between iron and iodine in DFS. All the coating materials formed an effective physical barrier between iron and iodine in the salt. Even after one year of storage, we only observed a 30 per cent loss of iodine in one of the salt samples (formulated with premix coated with HPMC) at 45 °C; less than 10 per cent of the iodine added was lost in all the salts after 6 months of storage. At 45 °C and one year, having soy stearin as the outer coat was marginally better than having HPMC as the outer coat. The result supported our initial hypothesis that having 5 per cent HPMC and 5 per cent soy stearin may be as good as having a 10 per cent soy stearin coat in terms of iodine stability in the salt (Figure 5). Thus proving our hypothesis, the hydrophobic nature of the soy stearin contributed to the floating of the iron premix in the DFS.

More importantly, the comparable stability of iodine in the DFS formulated with iron premix made in the laboratory and the pilot plant, the consumer acceptance of the salt, further improvement of the technology, and consequent improvement of the iron status in the iron-deficient population that consumed the salt is a pointer to a successful scale-up of the technology and the impact that multiple micronutrient fortification of salt can have on a population (Diosady *et al.*, 2019; Jadhav *et al.*, 2019; Larson *et al.*, 2021; von Grafenstein *et al.*, 2021). Therefore, this has encouraged adding more micronutrients to DFS, such as folic acid, vitamin B12, and zinc (Modupe *et al.*, 2019, 2021; Modupe, 2020; Modupe and Diosady, 2021).

Conclusion

The study explores the underlying reasons for the floating of iron premix in the DFS and provides solutions to address this problem. Different coating formulations were studied to optimize the best coating formulations in the laboratory and further validated from laboratory scale to pilot scale-up industrial trials. It was found that using 25–30 per cent (in weight) titanium dioxide for color masking supports the iron premix's effective coating without negatively impacting its color. On the other hand, the hydrophobic nature of soy stearin with the use of 10 per cent or more for coating caused the iron premix to float in water. Double coating of the iron premix with 5 per cent HPMC and 5 per cent soy stearin solved the floating problem experienced during field trials in India. About 90 per cent iodine in the DFS formulated with the premix was retained after 6 months of storage. This report provides an overview of the scale-up process and the influence of different unit operations during scale-up to get the desired product quality. Thus, the dual coating approach was selected as the solution to prevent floating of the iron premix. This advancement has significantly contributed to the successful implementation of the salt fortification program in

India and its expansion to other countries, as evident in the acceptance of the salt in the populations in which the program has been introduced without any reported negative effect.

Author Contributions

Oluwasegun Modupe, Kiruba Krishnaswamy, and Yao Olive Li designed and carried out the experiments and prepared this manuscript. Diosady Levente was the Principal Investigator who supervised the study and edited the manuscript.

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Conflict of Interest

The authors declare no conflict of interest.

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