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Studies on the fortification of cane sugar with iron and ascorbic acid

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1. The feasibility of improving iron nutrition by fortifying cane sugar with Fe and ascorbic acid was studied.

2. It was found to be possible to add a number of Fe salts together with ascorbic acid to sugar without affecting its appearance or storage properties.

3. The absorption of Fe from fortified sugar eaten with maize-meal porridge or made into jam or biscuits was measured in ninety-four volunteer multiparous Indian women using the ^{59}Fe erythrocyte utilization method.

4. The absorption of Fe from sugar fortified with ascorbic acid and ferrous sulphate and eaten with maize-meal porridge was increased about twofold if the ratio, ascorbic acid:Fe was 10:1 by weight. If the ratio was increased to 20:1, Fe absorption was increased a further threefold.

5. Sugar fortified with soluble Fe salts, including $FeSO_4.7H_2O$, discoloured both tea and coffee; sugar fortified with ferric orthophosphate did not have this effect.

6. Fe from $FePO_4.H_2O$ was poorly absorbed when added with sugar to maize-meal porridge, and also when added with adequate quantities of ascorbic acid. This form of Fe was absorbed much less well than was the intrinsic Fe present in the maize.

7. When sugar fortified with $FePO_4.H_2O$ and ascorbic acid was added to maize-meal porridge before cooking or was made into jam there was a several-fold increase in the amount of Fe absorbed.

In many parts of the world, where the average diet is predominantly cereal-based, a major factor contributing to the high incidence of iron deficiency is the poor absorption of the Fe from such foodstuffs. In one study the geometrical mean values for Fe absorption from rice, maize and wheat were 1, 3 and 5% respectively (Martinez-Torres & Layrisse, 1973), and even for an Fe-deficient group of subjects the mean absorption rates from the same staples were only 3·5, 5·3 and 8·5% respectively (Sayers, Lynch, Jacobs, Charlton, Bothwell, Walker & Mayet, 1973; Sayers, Lynch, Charlton, Bothwell, Walker & Mayet, 1974*a*). These results underline

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the need for the fortification of these diets with Fe. However, for this type of programme to be effective it is necessary not only to find a suitable 'carrier' for the supplemental Fe but also to show that this Fe is satisfactorily absorbed. The results of food fortification studies have been disappointing (Elwood, 1968), and this can be ascribed to the fact that Fe added to cereal diets is subject to the same factors that inhibit the absorption of the native food Fe (Bjorn-Rasmussen, Hallberg & Walker, 1973; Layrisse, Martinez-Torres, Cook, Walker & Finch, 1973). In previous studies it has been found that these inhibitory factors can be overcome by the addition of agents which promote Fe absorption, e.g. ascorbic acid (Sayers *et al.* 1973). There is also evidence that common salt (NaCl) can be used as a 'carrier' for the addition of Fe and ascorbic acid (Sayers *et al.* 1974*a*; Sayers, Lynch, Charlton, Bothwell, Walker & Mayet, 1974*b*). However, most forms of Fe adversely affect the storage properties of NaCl, particularly the coarse varieties eaten in many parts of the world (Sayers *et al.* 1974*b*). For this reason a study was done to find out whether cane sugar, which is widely and fairly uniformly eaten, might represent a more suitable 'carrier' for Fe and ascorbic acid.

EXPERIMENTAL

Preparation of fortified sugar

Fe was added to commercial white cane sugar (Huletts Sugar Refining Co. Ltd, Durban, South Africa) at two levels of fortification, either 100 or 200 mg/kg, in the form of a number of white or lightly coloured Fe compounds. These included ferrous sulphate, ferrous ammonium sulphate, ferric ammonium sulphate, (all purchased from BDH Chemicals Ltd, Poole, Dorset, UK), ferric sulphate, ferric nitrate, ferric orthophosphate (all purchased from Riedel-de Haen, Seelze-Hanover, West Germany), ferric glycerophosphate (E. Merck AG, Darmstadt, West Germany), ferric fructose (prepared by the method of Charley, Sartar, Stitt & Saltman (1963)) and ferric sodium EDTA (prepared by the method of Sawyer & McKinnie (1960)). In some experiments ascorbic acid (L(+)), analytical grade (BDH Chemicals Ltd, Poole, Dorset), was also added to the sugar to produce a concentration of either 1 or 2 g/kg. Supplementation was carried out in two different ways: either the Fe, and where applicable the ascorbic acid, was dissolved in distilled water and then sprayed onto the sugar, or the sugar was dampened with water (1 g/kg) before mixing with each of the finely ground, dry supplements in turn. In each instance the sugar was dried thoroughly after supplementation, using warm air.

Samples of fortified sugar were kept for up to 2 years in a laboratory in which the temperature was 22–27° and the mean relative humidity was 55%. Other samples were kept in the Huletts Sugar Refinery in Durban, which lies in a subtropical zone where the temperature ranged between 6 and 35° and the mean relative humidity was 77%. The standard quality-control method employed by the Huletts Sugar Refining Co. Ltd was used to assess consumer acceptability. The extinction at 420 nm of a 500 g/l solution in distilled water, measured using a spectrophotometer (Beckman Instrument Corp., Fullerton, California, USA), was multiplied by 163, and the sample was judged to be acceptable if the value obtained was less than 50. The ascorbic acid content after storage was measured using the method of Roe (1954). As sucrose may lead to spuriously high extinction values with this method a sucrose blank was used in all estimations.

Absorption studies

Ninety-four multiparous housewives aged between 30 and 50 years (mean 39 years) took part in the study. They belonged to a low socio-economic group and lived in a municipal housing scheme at Chatsworth, near Durban. All were of Indian descent. It has previously been established that Fe deficiency is a common problem in this community (Mayet, Adams, Moodley, Kleber & Cooper, 1972).

The absorption of Fe from maize-meal porridge, apple jam with bread, and wheat biscuits was measured. In every instance the meal eaten by each subject contained 20 g white sugar fortified with Fe and also, sometimes, with ascorbic acid. In most of the experiments two meals were eaten on successive mornings after an overnight fast, the Fe in the fortified sugar was labelled for one meal with 2·5 μCi ^{59}Fe and for the other meal with 2·5 μCi ^{55}Fe (Radiochemical Centre, Amersham, Berks., England). In two of the studies, however, a single meal of maize-meal porridge was eaten: in this instance the Fe in the fortified sugar was labelled as before with ^{59}Fe, but the intrinsic maize Fe was labelled with ^{55}Fe by hydroponic culture (Hussain, Walker, Layrisse, Clark & Finch, 1965), and sufficient was mixed with unlabelled maize meal to supply 2·5 μCi/subject. Each subject took part in one experiment only.

In all experiments no food or drink was allowed for 4 h after the test meal had been eaten. After 2 weeks the subjects were again assembled after an overnight fast and samples of blood were taken for the determination of ^{59}Fe, ^{55}Fe, haemoglobin, serum Fe concentrations and the unsaturated Fe-binding capacity. Each subject then drank 50 ml of a solution containing 30 mg ascorbic acid and 3 mg Fe as $FeSO_4.7H_2O$ labelled with 2·5 μCi ^{59}Fe; no food or drink was allowed for the subsequent 4 h period. A further 2 weeks later samples of blood were again obtained and the ^{59}Fe concentrations determined. The absorption of the Fe in the solution, which was then calculated by difference, provided an index of each individual's absorbing capacity.

Preparation of meals

Maize-meal porridge. Sufficient maize meal was weighed out to provide 30 g dry maize/subject. It was cooked in water (1 g maize meal/4 g water) for 20–25 min at 90–95°. The final weight of porridge eaten by each subject was approximately 100 g (plus 20 g fortified sugar). In one experiment the fortified sugar was added before cooking, but in the other experiments it was sprinkled on top of the porridge.

Apple jam. Apples were peeled and cored and then homogenized using a Waring blender. The homogenate was boiled for 20 min, then the fortified sugar (670 g/kg) was added and the mixture was boiled for a further 20 min. The jam was eaten with a slice of commercially available white bread (made from flour of 70–80% extraction and weighing approximately 90 g).

Biscuits. A biscuit was made for each subject by mixing 60 g white flour (70–80% extraction) with baking powder, salt, 20 g butter and 20 g fortified sugar. After thorough kneading to ensure even distribution of the isotope, the biscuits were baked for 40 min at 190°.

Radioisotopic, statistical and chemical methods

Blood samples (10 ml) and portions of standard Fe solutions and foods were prepared for differential radioactive counting by the method of Katz, Zoukis, Hart & Dern (1964). The quantities of ^{55}Fe and ^{59}Fe in the processed samples were determined using a liquid scintillation system (Insta-Gel; Packard Instrument Co., Downers Grove, Illinois) and a Tri-Carb AAA spectrometer (Model 3375; Packard Instrument Co.), which automatically adjusted for quenching. The counting efficiency at optimal gain and window settings was 24% for ^{55}Fe and 42% for ^{59}Fe. The ^{59}Fe activity in the 4 ml blood samples collected immediately before the 'reference Fe salt' was administered and 2 weeks later was assessed with reference to suitable standards, using an Auto-Gamma Tri-Carb spectrometer (Model 3001; Packard Instrument Co.). All absorption values were calculated on the basis that 100% of the absorbed radioactivity was present in the haemoglobin of circulating erythrocytes, and that the blood volume for each subject was 65 ml/kg body-weight. The significance of differences between the absorptions of the two isotopes was assessed using Student's t test for paired observations.

Serum Fe concentrations were estimated using a modification (Bothwell & Finch, 1962) of the method of Bothwell & Mallett (1955) in which sulphonated bathophenanthroline was used as the colour reagent. The unsaturated Fe-binding capacity was determined by the method of Herbert, Gottlieb & Lau (1967). The Fe content of digested samples of food was estimated using a modification (Bothwell & Finch, 1962) of the method of Lorber (1927).

Ethical considerations

Before making this study, we obtained approval from the Committee for Research on Human Subjects of the Faculty of Medicine, University of the Witwatersrand. Written consent was obtained from all subjects after the nature of the study had been explained to them. In relation to the radiation dosage, we calculated that if the whole of each test dose had been retained, the total radiation dose averaged over a period of 13 weeks would have been approximately 20% of the permissible whole-body burden for continuous exposure in the instance of ^{59}Fe and 0·2% in the instance of ^{55}Fe (International Commission for Radiation Protection, 1960).

RESULTS

Preparation of fortified sugar

Fortification by spraying a solution of any of the soluble Fe compounds, with or without ascorbic acid, onto dry sugar was unsuccessful as a purple colour developed within minutes. Over a period of a few days the colour changed to brown, and this was accompanied by oxidation of the ascorbic acid so that only 30% of the initial reduced ascorbic acid content remained after 48 h. The alternative method proved much more successful. No colour change or loss of reduced ascorbic acid content was found during storage for 2 years, even under subtropical conditions, if the dry, powdered supplements were mixed with dampened sugar crystals. Although the appearance of sugar fortified by this method with any of the Fe compounds and ascorbic acid was subtly different when directly compared with unsupplemented sugar, it was nevertheless considered to be acceptable to consumers, as it passed the standard quality control test of the sugar-refining company.

As there did not appear to be any obvious differences between the appearances and 'shelf-life' of sugar samples fortified with the various Fe compounds, it was initially decided to restrict the absorption studies to sugar fortified with $FeSO_4.7H_2O$, with or without ascorbic acid.

Absorption of Fe from maize-meal porridge and sugar fortified with $FeSO_4.7H_2O$ or $FeSO_4.7H_2O$ and ascorbic acid

In the first experiment ten subjects ate maize-meal porridge intrinsically labelled with ^{55}Fe. Sugar (20 g, fortified with 2 mg Fe as $^{59}FeSO_4.7H_2O$ and 20 mg ascorbic acid) was sprinkled over the porridge and eaten at the same time (Table 1). There was no significant difference (t 1·01, $P < 0·4$) between the absorption of the intrinsic Fe (mean 10·5%, SD 6·0) and that of the extrinsic Fe (mean 10·1%, SD 6·7). This finding confirmed the considerable amount of reported evidence which indicated that the non-haem Fe in a meal is absorbed from a common pool (Bjorn-Rasmussen, Hallberg & Walker, 1972; Cook, Layrisse, Martinez-Torres, Walker, Monsen & Finch, 1972; Sayers *et al.* 1973), and established the validity of using an extrinsic label only to assess Fe absorption in subsequent studies.

In the second experiment maize-meal porridge was eaten on two successive mornings by ten subjects (Table 2). On one morning fortified sugar containing 2 mg Fe as $FeSO_4.7H_2O$ was added to the porridge and on the second morning the sugar also contained 20 mg ascorbic acid; the mean absorption values were 3·8% (SD 2·5) and 6·9% (SD 4·3) respectively; the difference was significant (t 2·33, $P < 0·05$). A similar experiment was then done with eleven subjects for whom the doses of Fe and of ascorbic acid were increased to 4 mg and 40 mg respectively. Again there was evidence that absorption of Fe was increased in the presence of ascorbic acid (Table 2). The mean percentage absorption when the sugar was supplemented with both ascorbic acid and iron was 10·3% (SD 6·9) compared with 6·2% (SD 4·8) when it contained only Fe; the difference was significant (t 2·34, $P < 0·05$).

Table 1. *Absorption values for intrinsic iron (^{55}Fe)* and extrinsic Fe (2 mg given as $^{59}FePO_4.H_2O$, or $^{59}FeSO_4.7H_2O$) from a meal of maize-meal porridge† given with Fe- and ascorbic acid (AA)-fortified sugar‡, to female subjects*

(Mean values and standard deviations; no. of subjects in parentheses)

Fe Supplement	Final Fe content of meal (mg)	AA supplement (mg)	Haemoglobin (g/l)		Plasma Fe (mg/l)		Percentage saturation of total Fe-binding capacity		Fe absorption (%): Reference salt		Fe absorption (%): Intrinsic ^{55}Fe		Fe absorption (%): Extrinsic ^{59}Fe		Ratio, extrinsic Fe: intrinsic Fe	
			Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
$^{59}FeSO_4.7H_2O$ (10)	3·0	20	116	34	0·69	0·32	16·0	7·7	66·2	29·9	10·5	6·0	10·1	6·7	0·9	0·1
$^{59}FePO_4.H_2O$ (7)	3·2	40	129	14	1·13	0·47	29·1	13·5	33·3	13·3	9·4	9·5	1·4	1·5	7·1	2·4

* Maize was grown hydroponically and labelled intrinsically with ^{55}Fe (Hussain, Walker, Layrisse, Clark & Finch, 1965).
† Approximately 100 g porridge/subject, after 30 g dry maize meal had been cooked in water (1 g maize meal/4 g water) for 20–25 min at 90–95°.
‡ Dry, powdered supplement mixed with dampened sugar crystals (1 g water/kg sugar); 20 g/subject, sprinkled over the porridge.

Table 2. *Absorption values for iron from a meal of maize-meal porridge* given with 20 g sugar fortified† with $FeSO_4.7H_2O$ or $FePO_4.H_2O$, with or without additional ascorbic acid (AA) supplements, to female subjects on two successive mornings‡*

(Mean values and standard deviations; no. of subjects in parentheses)

Fe supplement	Final Fe content of meal (mg)	AA supplement (mg)	Haemoglobin (g/l)		Plasma Fe (mg/l)		Percentage saturation of total Fe-binding capacity		Fe absorption (%): Reference salt		Fe absorption (%): Without AA		Fe absorption (%): With AA	
			Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
$FeSO_4.7H_2O$ 2 mg (10)	3·6	20	107	30	0·85	0·49	18·8	11·2	45·2	27·6	3·8	2·5	6·9	4·3
4 mg (11)	5·2	40	130	8	0·92	0·34	20·2	8·3	38·6	30·0	6·2	4·8	10·3	6·9
$FePO_4.H_2O$ 2 mg (21)	3·4	40	118	15	1·01	0·58	22·2	12·1	31·9	25·5	1·3	1·7	2·9	2·9

* Approximately 100 g porridge/subject, after 30 g dry maize meal had been cooked in water (1 g maize meal/4 g water) for 20–25 min at 90–95°.
† One morning the sugar was fortified with the Fe salt only, the second morning AA was added.
‡ Dry, powdered supplement mixed with dampened sugar crystals (1 g water/kg sugar); 20 g/subject, sprinkled over the porridge.

While it was clear from these experiments that ascorbic acid was increasing the absorption of Fe, the extent of the increase was less than that which we had previously found in studies using fortified salt (Sayers *et al.* 1974*b*). A further study was therefore done in which the ratio, ascorbic acid: Fe was increased from 10:1 to 20:1. Nine subjects (mean values: haemoglobin content 116 g/l (SD 10), plasma Fe concentration 0·96 mg/l (SD 0·64), percentage saturation of total Fe-binding capacity 22·0 (SD 12·4)) were given the standard meal of maize-meal porridge; on one morning the sugar contained 2 mg Fe as $FeSO_4.7H_2O$ and 20 mg ascorbic acid, on the second morning the sugar contained the same Fe dose and 40 mg ascorbic acid. The larger dose of ascorbic acid was associated with an increase in the mean absorption of Fe from 5·7% (SD 5·5) to 19·6% (SD 12·6); this difference was highly significant (t 3·45, $P < 0·01$). The mean absorption of the reference Fe salt was 38·9% (SD 20·0).

Effect of $FeSO_4.7H_2O$-fortified sugar on tea and coffee

A series of studies was planned using Fe-fortified sugar in tea and coffee. However, it was discovered that a marked black discoloration developed almost immediately after the addition of the sugar to tea. The appearance was quite unacceptable whether the tea contained milk or not, and was similar whatever Fe compound was used to fortify the sugar, with the single exception of the insoluble $FePO_4.H_2O$. The addition of sugar fortified with ascorbic acid alone produced some lightening of the colour of tea without milk, reminiscent of 'lemon tea'. The effect was not obvious in the instance of tea with milk. The same pattern of findings was noted with coffee, but because coffee is darker than tea the change in appearance was far less striking.

These findings appeared to rule out sugar as a 'carrier' for dietary supplementation with soluble salts of Fe. However, the possibility remained that $FePO_4.H_2O$ might still prove suitable. Although very poorly absorbed under most conditions, it was found during a previous study in this laboratory that absorption from meals cooked with household salt (NaCl) fortified with both $FePO_4.H_2O$ and ascorbic acid was as good as when $FeSO_4.7H_2O$ was substituted for the $FePO_4.H_2O$ (Sayers *et al.* 1974*b*). A further series of experiments was therefore planned to examine the feasibility of fortifying sugar with $FePO_4.H_2O$ and ascorbic acid.

Absorption of Fe from maize-meal porridge and sugar fortified with $FePO_4.H_2O$, or $FePO_4.H_2O$ and ascorbic acid

On successive mornings twelve subjects were given a meal of maize-meal porridge with 20 g Fe-fortified sugar. On one morning the sugar contained 2 mg Fe as $FePO_4.H_2O$ and on the second morning, 40 mg ascorbic acid was also present. The mean percentage absorption rates were low on both occasions, being 2·1% and 4·1% respectively. Since these values were lower than had been obtained in any of the previous studies the experiment was repeated with a further nine subjects, with essentially similar results. When the twenty-one values were pooled together, the mean absorption without ascorbic acid was 1·3% (SD 1·7) as compared with

2·9% (SD 2·9) in the presence of ascorbic acid (Table 2). The difference was significant (t 3·59, $P < 0·01$). These low values contrasted with the mean absorption of the reference Fe salt, which was 31·9% (SD 25·5).

The question then arose as to whether the low absorption values obtained for these experiments also reflected the absorption of the intrinsic Fe present in the maize. A further study was therefore done in which seven subjects ate porridge made from ^{55}Fe-labelled maize onto which sugar containing 2 mg Fe as $^{59}FePO_4.H_2O$ and 40 mg ascorbic acid had been sprinkled. There was a significant difference (t 5·08, $P < 0·01$) between the mean absorption of the intrinsic Fe (9·4%; SD 9·5) and that of the added Fe (1·4%; SD 1·5) (Table 1).

In seeking a reason for the marked discrepancies between the present results and those reported previously by us for studies using NaCl fortified with $FePO_4.H_2O$ and ascorbic acid (Sayers *et al.* 1974*b*), one important difference was noted. In the previous studies the NaCl was added prior to the cooking of maize porridge or rice, while in the present studies the sugar was added only after cooking. This raised the possibility that heating was necessary for the formation of an absorbable Fe complex between the insoluble $FePO_4.H_2O$ and the ascorbic acid, and an experiment was done to study this possibility.

Effect of cooking on absorption of Fe in sugar fortified with $FePO_4.H_2O$ and ascorbic acid

A direct comparison between the effects of adding fortified sugar before and after the cooking of maize-meal porridge was then made using ten subjects (mean values for haemoglobin content 130 g/l (SD 12); plasma Fe concentration 0·71 mg/l (SD 0·25); percentage saturation of total Fe-binding capacity 17·0 (SD 7·6)). On one morning sugar containing 2 mg Fe as $FePO_4.H_2O$ and 40 mg ascorbic acid was sprinkled over the cooked porridge, while on the second morning the sugar was added before the porridge was cooked. The mean absorption rates were 1·8% (SD 1·1) and 12·7% (SD 9·3) respectively, a difference that was highly significant (t 3·79, $P < 0·01$). The mean absorption of the reference Fe salt was 44·1% (SD 22·4).

The effects of cooking on the absorbability of 2 mg Fe when given as $FePO_4.H_2O$ with 40 mg ascorbic acid was assessed using two other foods, jam and biscuits. In one study eight subjects were given jam and a slice of bread on two successive mornings. On one morning the jam had been prepared from fortified sugar containing $FePO_4.H_2O$ alone and on the second morning, both $FePO_4.H_2O$ and ascorbic acid were added. The presence of ascorbic acid was associated with a highly significant (t 3·91, $P < 0·01$) increase in Fe absorption, from a mean value of 2·3% (SD 1·5) to 13·8% (SD 8·5) (Table 3).

The effects of higher temperatures were assessed in a final experiment in which biscuits were baked using sugar fortified with $FePO_4.H_2O$ and ascorbic acid at the same levels as in the previous study. The mean absorption of Fe for eight subjects was 1·6% (SD 2·0) in the absence of ascorbic acid and 2·6% (SD 1·4) when ascorbic acid was present during baking; the difference was not significant (t 1·30, $P < 0·3$)

Table 3. *Absorption values for iron from a biscuit* or from apple jam* cooked with 20 g sugar fortified† with 2 mg Fe as $FePO_4.H_2O$ with or without 40 mg ascorbic acid (AA), and given to female subjects on two successive mornings‡*

(Mean values and standard deviations for groups of eight subjects)

	Fe content of meal (mg)	Haemoglobin (g/l)		Plasma Fe (mg/l)		Percentage saturation of total Fe-binding capacity		Fe absorption (%): Reference salt		Fe absorption (%): Without AA		Fe absorption (%): With AA	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Apple jam	2·3	131	11	1·00	0·34	23·7	10·0	47·2	22·1	2·3	1·5	13·8	8·5
Biscuit	2·4	130	16	0·87	0·35	23·8	10·1	57·3	28·0	1·6	2·0	2·6	1·4

* For details of preparation, see pp. 143–4.

† Dry, powdered supplement mixed with dampened sugar crystals (1 g water/kg sugar).

‡ One morning the food was prepared using sugar fortified with $FePO_4.H_2O$, the second morning the food was prepared using sugar fortified with $FePO_4.H_2O$ and AA.

(Table 3). The negative results were presumably ascribable to the destruction of ascorbic acid by the high temperatures that are necessary for baking (Sayers *et al.* 1973).

DISCUSSION

The great advantage of NaCl as a 'carrier' for dietary supplements is that it is consumed by all members of the population in relatively constant amounts which are related to the individual's total food intake (Sayers *et al.* 1974*a*). The consumption of sugar is certainly more variable, but a recent study revealed that most South Africans eat between 50 and 100 g daily, values similar to those reported for other countries (Walker, Holdsworth & Walker, 1971). In the present study sugar was found to be superior to NaCl as a 'carrier' for supplementation with Fe and ascorbic acid in one important respect; its consumer acceptability after storage in the hot, humid conditions which exist in many countries where nutritional Fe deficiency is an important problem. No colour changes developed under these conditions, whereas coarse, unrefined NaCl fortified with ascorbic acid and any soluble Fe compound became rapidly discoloured. Even when the supplemental Fe was in the form of the insoluble $FePO_4.H_2O$, discoloration remained a problem which was only partially solved by mixing the fortified NaCl with 25 g starch/kg (Sayers *et al.* 1974*b*).

Sugar also fulfilled the second essential requirement, that the supplementary Fe should be absorbed. When maize-meal porridge was eaten with fortified sugar containing $FeSO_4.7H_2O$ and ascorbic acid, both the intrinsic maize Fe and the supplemental Fe were absorbed to a similar extent. As has previously been found (Sayers *et al.* 1973; Bjorn-Rasmussen & Hallberg, 1974; Sayers *et al.* 1974*a*, *b*), the enhancing effect of the ascorbic acid was dose-dependent; only a modest effect was found when the ratio, Fe:ascorbic acid (by weight) was 1:10, but absorption was increased several-fold when it was 1:20. When the fortified sugar was added to tea, however, there was a marked colour change which would effectively rule out the use of sugar as a 'carrier' for dietary Fe supplementation in any country where tea is customarily drunk with sugar. For coffee, the same conclusion must probably be made, although the discoloration was less obtrusive, and it is conceivable that it might not be entirely unacceptable if the advantages of sugar as a 'carrier' proved compelling. The black colour is the result of the formation of iron tannates (Disler, Lynch, Charlton, Torrance, Bothwell, Walker & Mayet, 1975).

$FePO_4.H_2O$ did not discolour the beverages, and as it had already been found to be an effective substitute for $FeSO_4.7H_2O$ when the 'carrier' was NaCl (provided that ascorbic acid was also present), absorption studies were undertaken with sugar containing $FePO_4.H_2O$ and ascorbic acid. The absorption was very poor, and this was in striking contrast to the results obtained when NaCl was the 'carrier' (Sayers *et al.* 1974*b*). The reason for the discrepant findings was soon discovered: heat must be applied if the Fe in $FePO_4.H_2O$ is to be converted into an absorbable form by ascorbic acid. The fortified NaCl had been added to the porridge before it was cooked, and the fortified sugar after it was cooked; when the sugar was cooked with the porridge, absorption was markedly enhanced and was comparable with that found previously with NaCl. $FePO_4.H_2O$ is therefore an appropriate Fe compound for dietary supplementation only when it is present, together with an adequate amount of ascorbic acid, in the food before cooking. Most of the NaCl which is eaten each day is added during cooking rather than at the table: however, most sugar is probably consumed uncooked, although a proportion (which will obviously vary according to local custom) is of course used for such purposes as making jam.

The present findings indicate that there are real problems in using sugar as a 'carrier' for fortification of the diet with Fe and ascorbic acid. Soluble Fe salts that are well absorbed cause discoloration of tea and coffee, while insoluble salts are poorly absorbed unless added prior to cooking. These largely negative results underline the need for fresh approaches. The Fe content of some cereal diets seems adequate (Ramalingaswami & Patwardhan, 1949) but little of the Fe is absorbed (Martinez-Torres & Layrisse, 1973). In these circumstances the addition of ascorbic acid alone to a 'carrier' such as sugar may be all that is necessary to improve Fe nutrition. However, for these diets a significant proportion of the Fe content probably represents contamination. The extent to which the absorption of this contaminating Fe will be enhanced by ascorbic acid obviously depends on its nature. In this context the present findings may have relevance, as they indicate that a compound such as $FePO_4.H_2O$, unlike more soluble Fe salts (Bjorn-Rasmussen *et al.* 1972; Cook *et al.* 1972; Sayers *et al.* 1973), does not form a common pool with the intrinsic non-haem Fe in food, and is not affected to any extent by enhancing agents such as ascorbic acid. There is therefore no assurance that ascorbic acid would increase the absorption of dietary Fe derived from soil or other forms of contamination.

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