Factors Affecting the Bioavailability of Dietary Iron

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Iron is efficiently recycled in the body and little is excreted because its function in proteins of oxygen and electron transport is essential to life. Primary responsibility for maintaining adequate levels of this nutrient rests in an intestinal absorptive system that is attuned to the changing needs of the individual. The amount of iron absorbed from food is indirectly related to iron status. As iron stores fall, absorption increases, and once stores are replete, absorption falls to a low level. The few milligrams of iron necessary to replace daily losses must be absorbed from a diet that, in Western countries, usually contains a severalfold excess of this element. The fact that iron deficiency anemia is still a common condition indicates that not all of the iron present in this diet is capable of being absorbed.

Sources of Dietary Iron

Both the source and chemical form of dietary iron can markedly affect its availability for absorption. A mixed Western diet contains four sources of iron: food of animal origin, food of vegetable origin, fortification iron, and contamination iron (Table 1). For most of the world’s population, animal-derived foods are not available, and fortification iron is not yet widely distributed. In the vegetable category, the staples rice, maize, wheat, and beans have either moderate or poor iron availability. Therefore, it is not surprising that iron deficiency is prevalent among populations subsisting solely on these foods.

The factors that affect bioavailability of dietary iron will be presented here primarily in the context of a Western diet in which the amount of iron absorbed is not limited by the total iron content of the diet.

There are two major chemical forms of iron in a mixed diet, and each is absorbed by a different mechanism. Heme, containing iron in a porphyrin ring structure (see line art), is found in hemoglobin and myoglobin and accounts for nearly 40% of the iron present in animal tissue, including fish and poultry. Because of its unique absorptive mechanism and solubility at elevated pH, heme iron availability is unaffected by dietary components that either enhance or inhibit the availability of the nonheme iron present in the rest of the diet.

Nonheme iron is present in foods of vegetable origin and also accounts for the remaining 60% of iron in animal tissue. To be absorbed, nonheme iron must reach
the upper small intestinal mucosa in a soluble form.

Other dietary constituents aid or interfere with nonheme iron solubility and, thus, absorption. For example, although nonheme iron in meat is generally well absorbed, purified ferritin has low bioavailability when added to a vegetable meal (9). This may be an artifact due to chemical alteration during purification; however, it more likely demonstrates that meat digestive products are needed to facilitate ferritin absorption.

Other sources of nonheme iron are compounds added to fortify the diet with additional iron above its endogenous level. The most common sources are soluble iron salts or small-particle elemental iron. When taken without food, ferrous salts are better absorbed than ferric forms (6). This is probably related to the fact that ferric iron is insoluble in aqueous solution above pH 3, whereas the majority of ferrous iron remains soluble at pH 8.

Particle size is an important determinant of elemental iron solubility and availability for absorption. However, electrolytic iron has a higher bioavailability than hydrogen-reduced iron of the same particle size, probably due to the greater surface area of its dendritic-crystal structure (7).

Solubility alone cannot accurately predict bioavailability of different iron compounds used for fortification of wheat flour. Four soluble forms of iron were added to dinner rolls, and iron absorption was measured in human subjects (8). Reduced iron was absorbed the same as ferrous sulfate, but ferric orthophosphate and sodium iron pyrophosphate were only about 30 and 50% as available, respectively (8).

The iron content of the diet also can be increased by contamination during harvesting, processing, cooking, or storing food. Under the acidic conditions occurring in fermentation of maize and sorghum beer, iron was extracted from cast-iron drums and was shown to be well absorbed from the beer (9). At the other extreme, soil contamination of teff when it is threshed under the hooves of cattle was reported to account for most of the 200-300 mg daily iron intake of Ethiopians (10). Oxides and hydroxides of iron, common insoluble environmental forms, do not exchange with soluble radiiron salts used to measure iron absorption in humans. Therefore, the actual nutritional value of contaminant iron cannot be directly determined (11).

Predicting the Bioavailability of Dietary Iron

Methods for predicting availability of dietary iron in vitro have concentrated on duplicating the chemical environment of the stomach and upper small intestine. Although use of human gastric juice has been reported, more often food is incubated with hydrochloric acid containing pepsin at 37°C (12). After adjusting the solution to pH 7.5, the fraction of iron that was ionizable (reacting with α,α-dipyridyl) was found to correlate well with percentage iron absorbed in human studies; soluble iron, containing complexed as well as ionizable iron, did not (13). Other techniques determine the proportion of ionic iron in the ferric or ferrous state, thus providing a useful monitor of chemical changes occurring during food processing (14).

Although the amount of dietary iron that is chemically suited for absorption can be determined using in vitro techniques, animal studies are necessary to measure the actual absorption and utilization of food iron. Growing animals, depleted of iron stores through phlebotomy or by consuming an iron-deficient diet, are fed a diet containing graded levels of iron provided by a food item or an iron salt. The hemoglobin, hematocrit, or body weight response to this added iron is then compared to the response observed in a comparable group of animals fed identical levels of iron from highly available ferrous sulfate. In this manner the biological value of the iron contained in the test ingredient can be calculated relative to ferrous sulfate (15). Radioisotope retention by animals fed meals labeled with radiiron is another method that has been used and validated by showing that extrinsically added iron exchanges completely with iron in the diet (16, 17).

Iron bioavailability studies conducted in rats have provided much useful information concerning foods and iron compounds that are good or poor sources of iron. However, using animals to assess food interactions with dietary iron and to estimate its bioavailability in a complex meal is more difficult. Recent data suggest that a primar model may prove useful for these purposes (18).

The very small amount of iron absorbed relative to the iron content of the diet makes chemical iron balance studies in humans very difficult. Any analytical or sampling error may mask the effect on iron balance attributed to the diet.

Direct measurement of iron absorption using minute quantities of radioisotopes of iron has generated the most meaningful data on iron absorption in humans. Absorption is determined two weeks after a test meal by either measuring erythrocyte incorporation or whole-body retention of radioiron (4). Although at first radioiron was biologically incorporated into the food under study, it was subsequently found that a soluble form of inorganic iron added extrinsically to a meal exchanges completely with other dietary nonheme iron released into a common pool of available iron in the acidic environment of the stomach (19, 20). The addition of biosynthetically tagged heme iron likewise labels that component of dietary iron, enabling the proportional absorption of each iron form in a complex meal to be determined (21).

In some studies, a standardized dose of ferrous sulfate and ascorbic acid is included as one test meal, the absorption of which is based solely on the level of iron stores in each subject (22). By expressing iron absorption from a test meal as a ratio to the amount of ferrous ascorbate absorbed in the same individual, intersubject variability due to differences in iron status can be minimized. Additionally, this ratio is useful in predicting the amount of iron that would be expected to be absorbed by people differing in iron status from those in the test population. Adjusting the data to a reference dose absorption of 40% provides an index of how much iron in the test meal would be absorbed by subjects without any iron stores (11).

### Dietary Components Affecting Bioavailability

Although an iron salt may exhibit high availability when fed alone, adding it to a meal such as maize, wheat, or black beans reduces the level of iron absorption considerably (19). Conversely, when a glass of orange juice is included in a breakfast meal, iron absorption can be
increased 2½ times (23).

These examples demonstrate that iron absorption from a complex meal represents the summation of all enhancing and inhibiting components present in that meal. The iron absorption enhancers and inhibitors that have been studied most extensively in humans and their relative potencies are shown in Table II. Certain sugars, amino acids, and amines released during enzymatic digestion may improve iron availability by decreasing both the precipitation and polymerization of iron in aqueous solutions, probably by interfering with the formation of water bridges with iron molecules. Substances capable of forming chelates with iron promote absorption by maintaining the metal in a soluble, absorbable state (24).

Enhancers
Ascorbic Acid. When ascorbic acid is added to a meal, the increase in iron absorption is proportional to the dose, even at levels as high as 1 gm (26,27). Facilitation of iron absorption appears to operate in two ways. As a reducing agent, ascorbic acid maintains the normally ferric form of food iron in a more soluble ferrous state. Secondly, under acidic conditions in the stomach, ascorbic acid forms a complex with iron that remains soluble as the pH increases in the upper small intestine (28).

The addition of orange juice containing 40–50 mg ascorbic acid to a breakfast meal of bread, egg, and tea or coffee was found to increase iron absorption from 3.7 to 10.4% (29). Ascorbic acid also was shown to account for the sevenfold increase in iron absorption from maize porridge when papaya was included in the meal (30). Because oxidation and heating destroys much of the ascorbic acid in food during storage and cooking (31), the ascorbic acid content of raw foods may not accurately predict the degree of iron absorption enhancement expected from processed foods.

Animal Tissue. Besides contributing highly available heme iron, animal tissue in the diet produces a twofold to fourfold increase in the absorption of nonheme iron. This phenomenon applies to a variety of meat products, but eggs, milk, and cheese either have no effect or are somewhat inhibitory to iron absorption (32). The exact mechanism by which animal tissue promotes iron absorption is still unknown, but it has been suggested that amino acids and/or polypeptides arising from proteolytic digestion might chelate dietary nonheme iron, thereby facilitating its absorption.

Martinez-Torres and Layrisse observed a twofold increase in iron absorption when 100-g fish was added to a meal of black beans (33). They found this same magnitude of effect when the total amino acid content of the fish was added in purified form or when 210-mg cysteine (the amount present in 100-g fish) was added alone. Like ascorbic acid, cysteine also is a reducing agent.

Recently, studies by the same researchers have shown enhancement of iron absorption from maize, black beans, and soybeans when this level of cysteine was added after cooking the test meals (34).

Other workers have shown increased iron absorption when beef, fish, chicken, or calf thymus was added to a maize meal, but they noted no response for egg white cysteine, or a water extract of beef (35). They proposed that meat aids iron absorption by countering luminal factors that otherwise would interfere with the transport of iron to the mucosal cell surface.

Inhibitors
Much of the early work in iron nutrition focused on factors that would increase the amount of iron absorbed from the diet. Basal meals in these studies consisted of staple foods such as corn, beans, or rice, which, by themselves, exhibit low intrinsic iron bioavailability.

To better assess inhibitory factors in the diet, chemically defined meals were formulated using purified sources of protein, fat, and carbohydrate; ferric chloride was added to bring the iron content to 4 mg. Adding to this meal almost any nonmeat food that did not contain ascorbate usually resulted in a drop in iron absorption. The biochemical mechanism for this inhibition remains unclear in most instances. The following are the food constituents that have been most extensively studied in their effect on iron absorption in humans.

Tea. Disler and co-workers were the first to report the dramatic reduction in iron absorption when tea was drunk with a meal (36). A single cup of tea reduced absorption of ferric chloride from 22 to 6%, and even with a complex meal, tea caused iron absorption to fall from 11 to 2.5%. Tannins extracted from tea leaves by hot water were shown to account for this inhibition. Neither caffeine nor tannin-free tea had any effect on iron absorption in rats, whereas tannic acid or purified tannins extracted from tea produced the same degree of inhibition as brewed tea itself (37). This effect is probably due to formation of an insoluble iron-tannate complex in the gut that renders a significant proportion of dietary iron unavailable for absorption. So great is the inhibitory effect of tea, it has been proposed as one way to reduce dietary iron absorption in patients with iron overload diseases (38).

Coffee. Even in the presence of supplemental ascorbic acid, coffee as well as tea inhibits iron absorption (39). In our own studies, coffee has proved to be about two thirds as inhibitory as tea. When subjects drank coffee an hour after a meal, iron absorption was reduced the same amount as when coffee was taken with a meal. Greater inhibition was seen with stronger coffee.

Although the mechanism of the coffee effect has not been identified, polyphenolic compounds are present in coffee and could possibly act in the same manner as the chemically related tannins in tea. Evidence that tea and coffee may act differently on iron absorption has been presented by recent in vitro data. When added to a suspension of cooked pinto beans, tea reduced the amount of soluble iron by 88%. Coffee, on the other hand, did not decrease the amount of iron in solution but was responsible for oxidizing all to the ferric state (40).

Soy Products. Based on studies using human subjects, we have recently reported a substantial inhibition of iron absorption associated with soy products (41). When substituted for egg albumen as the protein source in a semi-purified diet, full-fat soy flour, textured soy flour, and isolated soy protein reduced iron absorption by 82, 65, and 92%, respectively. Similarly, a 50 to 60% decrease was observed when textured soy flour was added to ground beef in a hamburger meal. Follow-up studies showed that the inhibitory effect attributed to isolated soy protein could not be completely eliminated by extensive heating or by adding the enhancers ascorbic acid or meat to the meal. The precise mechanism behind this soy effect remains to be determined, but it appears to influence iron absorption to a greater degree when soy provides all of the

Table II. Food Components Affecting Dietary Iron Bioavailability

<table>
<thead>
<tr>
<th>Food Components</th>
<th>Potency</th>
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<tbody>
<tr>
<td>Enhancers</td>
<td>++</td>
</tr>
<tr>
<td>Ascorbic Acid</td>
<td>++</td>
</tr>
<tr>
<td>Animal Tissue</td>
<td>+++</td>
</tr>
<tr>
<td>Inhibitors</td>
<td></td>
</tr>
<tr>
<td>Tea</td>
<td>++</td>
</tr>
<tr>
<td>Soy Products</td>
<td>++</td>
</tr>
<tr>
<td>Coffee</td>
<td>++</td>
</tr>
<tr>
<td>EDTA</td>
<td>++</td>
</tr>
<tr>
<td>Calcium, Phosphate Salts</td>
<td>++</td>
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<tr>
<td>Eggs</td>
<td>++</td>
</tr>
<tr>
<td>Yolk</td>
<td>++</td>
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<tr>
<td>Albumen</td>
<td>++</td>
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<tr>
<td>Wheat Bran</td>
<td>++</td>
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<tr>
<td>Phytate</td>
<td>±</td>
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<td>Fiber</td>
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*When the major proportion of dietary protein is provided by soy.

The effects of EDTA on iron absorption appear to depend on both the absolute amount of EDTA in the diet as well as the molar ratio of EDTA to iron. When this molar ratio is less than 1, absorption from the meal may be enhanced, especially when basal availability of food iron is low.

Calcium/Phosphate. In animal studies, calcium and phosphate compounds have been reported to decrease nonheme iron availability (46,47). Monsen and Cook demonstrated that when a semi-purified meal was fed to humans, the addition of either calcium chloride or potassium phosphate alone did not affect iron absorption (48). However, when both were added simultaneously to the meal, iron absorption was reduced 53-73%. These results may be explained by the adsorption of iron onto the insoluble calcium phosphate complex formed in the alkaline environment of the small intestine, rendering it unavailable for absorption.

Eggs. Although eggs contain approximately 1 mg of iron, the iron is poorly available to humans and the egg yolk further inhibits absorption of other dietary iron (29). The addition of egg white to ferrous ascorbate reduced iron absorption from 33 to 14%, and added yolk dropped absorption even further to 3.9% (49). The vitellin fraction of yolk, high in phosphoprotein, was believed to account for this inhibition.

Cook and Monsen used a semi-purified diet containing egg albumen in human studies to assess the effect of various animal proteins on iron absorption. Powdered egg produced the same level of iron absorption as whole milk, cheese, or the unsubstituted basal diet, indicating that not all animal-derived protein enhances iron absorption (32). The same workers subsequently showed that doubling the quantity of egg albumen in the meal caused a drop in iron absorption from 2.3 to 1.4%. On the other hand, omitting it completely from the formulation increased iron absorption by nearly threefold (50). Taken together, these studies show that both the white and the yolk of hen’s eggs can inhibit the absorption of nonheme iron contributed by other foods eaten in the same meal.

Wheat Bran. In an early human study using chemical balance methods, Widdowson and McCance showed that more iron was absorbed from white bread than from brown bread, even though brown bread contained 50% more iron (51). More modern studies employing an extrinsic radioiron tag have demonstrated that iron absorption is reduced in direct proportion to the amount of bran added to a meal (52).

Because wheat bran is high in phytate, a compound known to bind iron and other trace minerals, this was proposed as the inhibitory compound in bran. More than half of the iron in wheat bran is present as monoferric phytate (53). In contrast to saturated ferric phytate, monoferric phytate remains soluble at neutral pH and its iron has been found to have relatively high bioavailability in rats, dogs, and man (54,55). However, only when purified sodium phytate is added to meals is iron absorption reduced; endogenous food phytate has no effect (51,56). In other human studies, iron absorption was diminished to the same extent when either dephytinized bran or whole bran was added to wheat flour muffins (55). Therefore, it was proposed that the inhibitory effect of bran was not necessarily due to phytate, but was perhaps associated with its high fiber content.

Fiber. In a recent review of the effects of fiber on trace mineral balance in man, Kelsay concluded that iron balance was unaffected by dietary fiber (57). However, a study in humans was conducted using dual radioiron tags added to two meals having identical nutrient composition but containing either 5.1 or 0.3 g crude fiber calculated from food tables. Iron absorption averaged 3% from the high-fiber meal and twice that level was absorbed from the low-fiber meal.

Further unpublished studies by the same group of investigators confirmed the bran inhibition of iron absorption but failed to show any effect due to pectin or purified cellulose.

Under in vitro conditions, iron reportedly was bound by acid detergent fiber and neutral detergent fiber from wheat and maize (58). Pectin, but not cellulose, also exhibited high iron binding activity in vitro and reduced by 40% the amount of iron absorbed by a group of iron-overloaded patients with hemochromatosis (59). Obviously, the effect of fiber on iron absorption has not been completely clarified. Studies using purified fiber sources may not be representative of the interaction of iron with foods that are naturally high in fiber.

Model for Estimating Iron Availability

A practical model has been proposed for estimating the amount of iron likely to be absorbed from any given diet (2).

The Authors

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Unpublished data.

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Literature Cited


