

# **Enhancing Iron Bioavailability of Vegetables through Proper Preparation—Principles and Applications**

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## **ABSTRACT**

Iron deficiency anemia is the most prevalent nutritional problem in the world today. AVRDC— the World Vegetable Center, an international, non-profit organization, aims to reduce malnutrition and poverty among the poor through vegetable research and development. Iron bioavailability of vegetables and food preparation ways to increase available iron in plants were investigated under the AVRDC Nutrition Program. Principles derived from iron bioavailability studies have been applied to design high-iron recipes based on conventional food preparations for iron-deficient areas, including southern and northern India. Enhancing nutrient bioavailability through optimum food preparations and legume-based recipe designs were effective in increasing available iron intake of schoolchildren in India. High-iron recipes including staple food and African indigenous vegetables based on East African countries are presently under investigation. This paper summarizes the major findings of iron bioavailability studies conducted mainly by the AVRDC and the applications of these studies towards improving iron nutrition in developing countries through international cooperation.

**Key words:** food-based nutrition, iron bioavailability, mungbean, recipes, tomato, vegetables

## Introduction

Iron deficiency is the most common nutritional disorder in the world and affects more than one billion people, particularly reproductive-age women and pre-school children in tropical and sub-tropical zones (ACC/SCN, 2000). It also has a serious impact on school-age children and working males. If uncorrected, iron deficiency leads to anemia of increasing severity, reduced work capacity, diminished learning ability, increased susceptibility to infection, and greater risk of death associated with pregnancy and childbirth (ACC/SCN, 1998).

Iron deficiency results from diets lacking in iron, reduced iron bioavailability, increased iron requirements due to pregnancy, and losses due to parasitic infections. Absorption of plant-based iron, though variable, is considered lower than that of iron from meat and it is greatly influenced by interactions with enhancers and inhibitors (Layrissse *et al.*, 1969; Cook, 1983). Populations in developing countries with limited resources avoid hunger by consuming more plant-based food than expensive animal based products (Baker and Mayer, 1979; World bank, 1994). The number of vegetarians is increasing worldwide, although their total iron intake may meet dietary recommendations (FAO/WHO, 1998), iron deficiency is common among vegetarians due to the low bioavailability of plant iron (Craig, 1994). Many nutrition programs aimed at decreasing

iron deficiency utilize supplements and/or fortification of diets (Scrimshaw, 1996). An alternative and sustainable approach would be to improve iron bioavailability of plant-based diets.

Food processing and iron chemistry are important factors affecting iron bioavailability. The chemistry of iron, particularly its valence, solubility, and types of chelation, influence its absorption. Food processing methods, such as baking, canning, drying, and cooking can have different effects on iron bioavailability, and therefore should be considered. Current methods for estimating iron bioavailability (IB) include animal bioassays, human assays, cell models and *in vitro* measurement. *In vitro* assay involves simulated gastrointestinal digestion using a commercially available enzyme and then measurement of the soluble iron released by this digestion to a dialysis tubing. Thus, this *in vitro* iron bioavailability assay is called iron dialyzability (ID) in this report. The *in vitro* measurement is simple, rapid, and inexpensive, and useful for food screening and identifying factors that might influence iron availability.

Vegetables provide multiple nutritional functions to human diets. Some are rich in micro-nutrients particularly  $\beta$ -carotene and iron; some provide macro-nutrients and energy (FIRDI/NPUST, 1998); while some are valued for health-promoting factors (Harborne, Baxter and Moss, 1999). In addition to their nutrient values,

vegetables nowadays are consumed worldwide to provide attractions and diversification in diet (Willtee, 1994). Higher consumption rate (vegetables and fruits, minimum 400 g and 5–9 servings a day) is recommended for health maintenance and cancer prevention (Steinmetz, 1996; Law and Morris, 1998).

Studies by Kapanidis and Lee (1995) at Rutgers State University, New Jersey, had indicated that *in vitro* bioavailability of iron in cruciferous vegetables can be enhanced from 5% to more than 20% through cooking. On the basis of that study, experiments were designed by the AVRDC and collaborators to (1) survey *in vitro* iron bioavailability of fruits and vegetables in raw and cooked forms and investigate better iron-source vegetables; (2) better understand the principles and mechanisms of cooking enhancing effects; (3) investigate household food preparation methods to increase available iron content in plant-based diets; (4) confirm through human study the enhancing ability of vegetables as assayed by the *in vitro* method; (5) design a legume-based diet with vegetables for higher iron consumption in South Asia.

The legume-base diet with vegetables was proved effective in improvement of nutrition status of school children. Higher iron recipe designs with nutritional studies of African indigenous vegetables and the previous iron bioavailability studies are currently included in a center-wise project, managed by

AVRDC–Regional Center for Africa in Tanzania, entitled “Promotion of Neglected Indigenous Vegetable Crops for Nutritional Health in Eastern and Southern Africa” funded by Federal Ministry for Economic Cooperation and Development (BMZ, Germany). In addition, this working model of the higher iron recipe design using African indigenous crops is being planned to apply in West African regions to promote iron consumption.

### The Principles

#### 1. Cooking enhancing effect on ID of vegetables and fruits

**1-1. ID of certain vegetables and fruits can be enhanced simply by boiling in water** (AVRDC, 1996 and 1997; Yang *et al.*, 1998; Yang, Tsou and Lee, 2002): Plant commodities commonly consumed in Asian diets, including various types of 46 vegetables (leaf, fruit, root, stem, flower, legume and mushroom), 16 fruits, and 2 cereals (rice and wheat) were measured for *in vitro* iron bioavailability in raw and cooked forms. Boiling in water for 10 min enhanced ID of 47 of the 64 of the tested commodities (Figure 1).

**1-2. Vegetables can be categorized into three groups based on their ID of raw and cooked form** (Yang, Tsou and Lee, 2002): Based on the ID values of uncooked and cooked, the 46 vegetables and 2 cereals could be

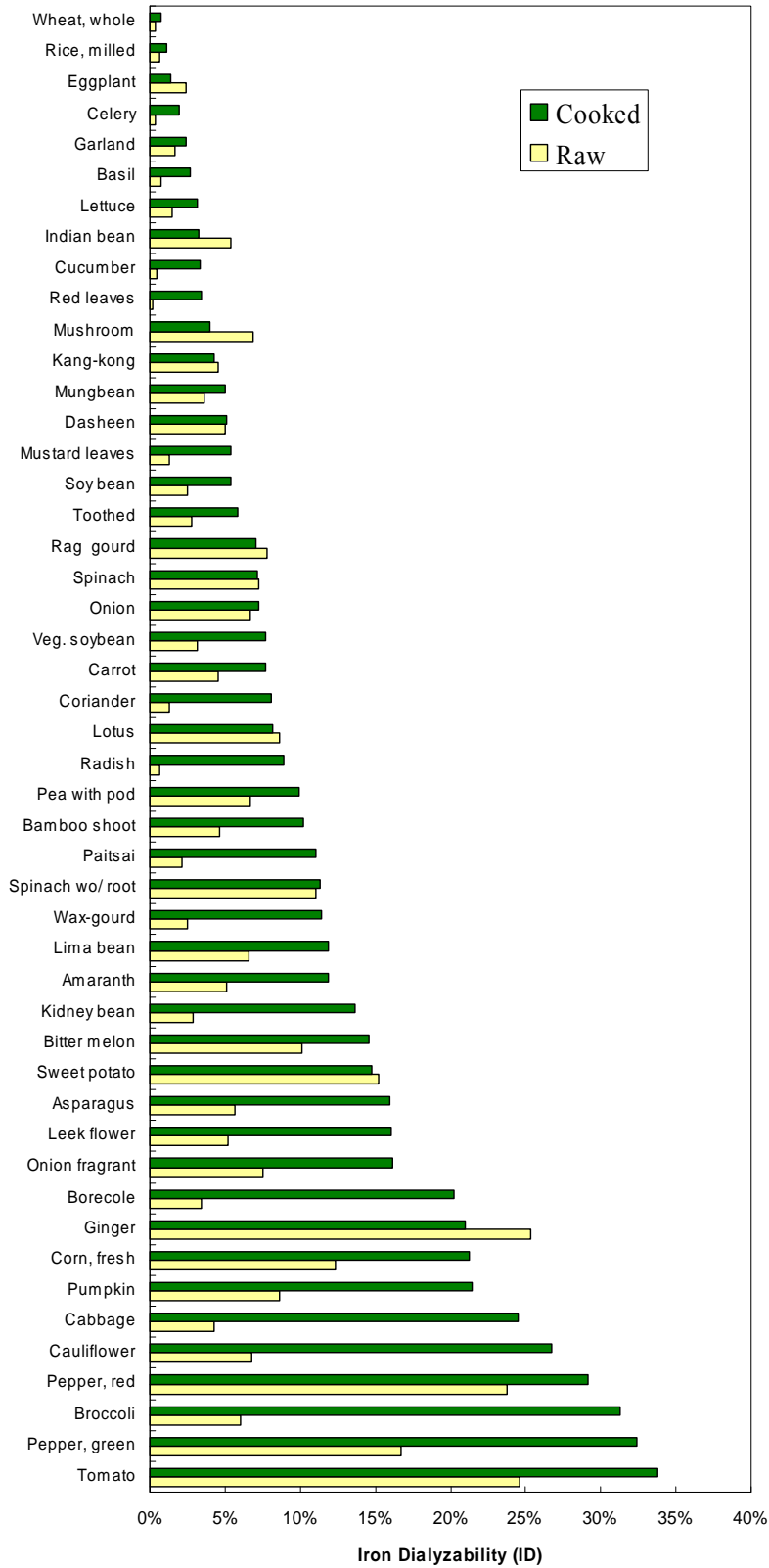


Figure 1. Iron Dialyzability (ID) of Raw and Cooked Vegetables

divided into three categories – Group 1: low ID in raw form but 2 times or more after cooking. Seventeen vegetables fall in Group 1, including cabbage, broccoli, cauliflower, amaranth and green kidney beans. Group 2: Low ID in raw form, ID slightly improved by cooking. A total of 24 items were in Group 2, including carrot, celery, eggplant, dried beans, kang-kong, onion, spinach, rice and wheat flour. Group 3: Relatively high ID (>10 %) in raw and cooked forms. Bitter melon, ginger, green and chili peppers, sweet potato and tomato were in this group 3.

**1-3. Enhancing effect of cooking is not as apparent in fruit as in vegetables** (AVRDC, 1997; Yang *et al.*, 1998): A wide variation in *in vitro* iron bioavailability, ranging from 0.6 to 21%, was measured among 16 selected fruits. An enhancing effect from cooking was observed in half of the samples. Cantaloupe, grapefruit, kiwi, and pineapple were found to have higher iron bioavailability compared to the others tested.

**1-4. Vegetables are a better iron source than fruits** (Yang and Tsou, 1998): ID of vegetables, legumes and cereals ranged from 0.2% to 25.3% in raw form and 0.8% – 33.8% in cooked items. Boiling vegetables in water raised available iron content

from an average of  $40.05 \pm 42.57 \mu\text{g}$  to  $75.03 \pm 53.37 \mu\text{g}$  per 100g of fresh weight. Boiling fruit produced a similar result, from  $32.29 \pm 25.06 \mu\text{g}/100 \text{g}$  to  $43.54 \pm 23.82 \mu\text{g}/100 \text{g}$ . In general, cooked vegetables are a better source of iron than fruits.

**1-5. Boiling fresh vegetables provides more available iron than processing, including pickling, blanching, canning, and drying** (AVRDC, 1996; Yang *et al.*, 1998): Selected processed vegetable products were measured for ID and compared to that of fresh form. Canned tomato paste and canned whole tomato fruit show very low iron availability compared to the fresh product. It might be due to lower bioavailability of the extrinsic iron from the can itself. Re-heated canned asparagus, bamboo shoots, and mushrooms could not increase ID. Pickling might provide more acid soluble iron, but no enhancing effect was observed through further cooking. Cooking tofu did not affect ID of its raw form. Hot-air drying at 80°C enhanced the iron bioavailability of raw cabbage, which has a cooking effect, but did not enhance spinach, which has no cooking effect. Further cooking the dried sample did not affect ID. Therefore, pickling, canning, blanching, and hot-air drying can enhance ID of raw materials. Double heating, like

cooking samples which had been blanched, hot-air dried, and canned was not able to increase ID of these processed products. There is only one enhancement in the first heat treatment. Cooking the material directly in boiling water for 10 minutes results in the most available iron.

**1-6. Enhancing effect of stir-frying is comparable to that of boiling**

(Yang *et al.*, 1998): Stir-frying with soybean oil is a common household cooking method for leafy vegetable dishes in Asia, and it showed an enhancing effect on tomato, cabbage, spinach, and cauliflower, and was found comparable to boiling. Stir-frying is an alternative method of preparation for vegetables, such as spinach, which did not show an enhancing effect by boiling.

**1-7. Cold storage suppresses ID of cooked vegetables**

(AVRDC, 1996): Storing raw tomato, spinach, and cabbage overnight at 4°C did not affect ID of uncooked form, nor did storing cooked samples for 3 hours at 4°C. Cold storage for more than one day was found to reduce available iron content of the cooked cabbage. This might be due to vitamin C oxidation and the slow reduction of available iron through interaction of the food matrix to form a complex.

**1-8. ID of mungbean can be improved by sprouting or by adding vitamin C before cooking**

(AVRDC, 1995; Yang *et al.*, 1998): The iron content in seeds and sprouts was found to be similar, but higher vitamin C and lower phytic acid contents were observed during mungbean sprouting process. Cooking slightly increased ID of mungbean seeds, but greatly increased ID of sprouts. Addition of vitamin C (more than 10-fold the iron content) could make iron in mungbean more available.

**2. Enhancing effect on staple foods and iron dense food through cooking with selected vegetables**

**2-1. Both raw and cooked tomato, and cooked cabbage and cauliflower exert an enhancing effect on an iron-dense food such as laver**

(Yang *et al.*, 1998): The iron content of laver (*Porphyra laciniata*) is as high as 957 ppm. Mixtures of cooked cauliflower and laver produced more available iron than was produced when the samples were cooked separately. This was also observed for cooking tomato and cabbage. Among these vegetables, tomato produced the most pronounced enhancing effect. Addition of raw tomato to cooked laver could increase available iron in the mixture. On the contrary, raw cabbage was found to be an inhibitor in a mixture of cooked laver and raw cabbage.

**2-2. Available iron of cooked mungbean is increased when mungbeans are cooked with tomato, cabbage and moringa** (AVRDC, 1998 and 2000; Yang *et al.*, 1998): Legumes are rich in protein. Often called “poor man’s meat,” they constitute an important supplement to the predominantly cereal-based diets in Asia. In vegetarian diets, or in diets containing low amounts of animal foods, they are an important source of protein. Mungbean was selected as a basic ingredient and cooked with other components to enhance the ID of the mungbean-based diet. Cooking with cabbage or tomato and mixing with raw tomato helps unlock the iron present in mungbean. Kale and sweet pepper showed no such enhancing effect.

**2-3. Cooking with tomato raises ID of soybean and lima bean** (AVRDC, 2000): Tomato was tested with other staple foods including sweet potato, rice, wheat flour, soybean and lima bean. ID enhancement was observed in soybean and lima bean when cooked with tomato.

### **3. Factors contributing to enhancing effects on ID in cooked vegetables**

**3-1. Mechanism of the cooking enhancing effect on cabbage** (AVRDC, 1999): A model was proposed to explain how cooking can enhance ID from cabbage – Iron in plant cells is stored mostly in

ferritin, from which iron may be released by proteases or denaturation by heat or low pH. Enzymes such as polyphenol oxidases are compartmentalized in cells until the cells are disrupted by blending or mastication. Almost immediately, the soluble iron becomes bound in iron-polyphenol complexes due to the action of polyphenol oxidases, and is thus rendered unavailable. Heating denatures the polyphenol oxidases preventing their action, but leaves intact a sufficient amount of ascorbic acid to maintain iron in a soluble form through chelation, even at pH 2 in the stomach and pH 6-7 in the intestine. Thus, more available iron can be absorbed from cooked cabbage than from the raw form.

**3-2. With/without and more/less iron enhancers and inhibitors explains the vegetable groups by cooking enhancing effect** (AVRDC, 2000): A strong iron chelator was used in the cooking effect study to explain that (1) Group 1 vegetables such as broccoli, cabbage, and mustard leaves contained both enhancers and heat related inhibitors. Cooking eliminates the inhibitors, and then enhancers raise up ID of cooked vegetables; (2) Group 2 vegetables such as kang-kong, mungbean and soybean lack both heat related inhibitors and enhancers. Thus no cooking enhancing effect was

performed and insufficient amount of enhancers to bring up ID; (3) Group 3 vegetables such as tomato and pepper contained less inhibitors and more enhancers. Lack of apparent cooking enhancing effect was an evidence of the lack of heat related inhibitors, and high ID in both raw and cooked forms explained the existence of enhancers.

## The Applications

**1. High-iron mungbean recipe design for South India** (Subramanian and Yang, 1998; AVRDC, 1999): High-iron mungbean recipes were designed based on the previous iron bioavailability studies and traditional food preparations in South India. The work was conducted through a joint effort by the researcher from India (Dr. M. A. Subramanian, Avinashilingam Deemed University, Coimbatore, India) and researchers from AVRDC, Taiwan.

Fifteen recipes were designed using mungbean dhal (dehulled split) and dehulled mungbean (dehulled split with hulls retained) as principal ingredient in combination with selected vegetables (Table 1). To enhance the bioavailability of iron, recipes should include one or more ingredients rich in ascorbic acid. Tomato mungbean dhal with rice was determined to be the best in terms of ID (11.28%). Mungbean dhal masial (mungbean dhal, tomato, onion)

ranked second (10.88%), and spinach koottu (mungbean dhal and spinach), and cabbage koottu (mungbean dhal and cabbage) ranked third and fourth with 10.6% and 10.1% iron bioavailability, respectively. The bioavailability of mungbean masial, which uses whole mungbean, recorded bioavailability of 8.83%, compared to 10.88% for the same recipe made with mungbean dhal. A salad made with soaked mungbean dhal or sprouts achieved the same percent iron bioavailability as mungbean (whole mungbean) masial. Recipes made only with rice and mungbean or mungbean powder were found to have low iron bioavailability. Recipes which included no enhancing ingredient had low ID.

**2. High-iron mungbean recipe design for North India** (Bains, Yang and Sundar, 2003): This work was conducted through a joint effort by the researcher from North India (Dr. Kiran Bains, Department of Food and Nutrition, Punjab Agricultural University, Punjab, India) and researchers from AVRDC, Taiwan. The high iron recipes were prepared to suit the palate of North Indians (Table 2). The selected ingredients were inexpensive and easily assessable to rural families as well as the urban poor.

**3. Enhancing bioavailability of iron from mungbean and its effects on health of schoolchildren:** The cooking enhancing effect could be extended to

**Table 1. High-Iron Mungbean Recipes for South India**

<i>Recipe name</i>	<i>Terms</i>	<i>Iron Dialyzability</i>
1. Mungbean <b>Masial</b>	▪ Cooked legumes and vegetables mixed or mashed	8.83%
2. Mungbean Dhal Masial		10.88%
3. Mungbean Dhal <b>Koottu</b> with Cabbage	▪ Mixed vegetable curry with coconut	10.10%
4. Dhal Koottu with <b>Drumstick leaves</b>	▪ <i>Moringa oleifera</i> , a high iron and vitamin A tree vegetable	5.63%
5. Mungbean Dhal Kootu with Amaranth		6.95%
6. Dhal Koottu with Spinach		10.59%
7. Tomato Rice with <b>Dhal</b>	▪ Dehulled split pulse	11.28%
8. Hot <b>Pongal</b>	▪ Harvest festival celebrated in Tamil Nadu in January which lends its name to two recipes in this book	5.76%
9. Sweet Pongal		5.08%
10. <b>Pesarattu</b>	▪ Mungbean dhal and rice pancake	9.33%
11. Tomato <b>Adai</b>	▪ Rice and pulses soaked, ground and cooked as a pancake with various ingredients for taste	9.71%
12. Salad		8.78%
13. <b>Pakoda</b>	▪ Small dough balls made from pulse and rice flour and onions, deep fried	5.06%
14. <b>Bonda</b>	▪ Large ball of legume flour dough deep fried	4%

Data source: Subramanian and Yang, 1998

low ID vegetables and legume by cooking together with high ID vegetables such as tomato and Moringa (AVRDC, 2000; Yang *et al.*, 2006). High iron mungbean recipes were designed accordingly for south and north India (Subramanian and Yang, 1998; Bains, Yang and Sundar, 2003).

Dishes selected and modified based on availability and prices of ingredients at the local markets were identified and used for the one year feeding trial conducted among 225 school children by Vijayalakshmi *et al.* (Avinashilingam Institute for Home Science and Higher Education for Women) in

**Table 2. High-Iron Mungbean Recipes for North India**

<i>Recipe name</i>	<i>Terms</i>	<i>Iron Dialyzability</i>
1. Dhuli Mung <b>Dhal</b>	▪ Preparations made of split dehulled or whole pulse	10.20%
2. <b>Sabat</b> Mung Dhal	▪ Whole	8.15%
3. <b>Parantha</b>	▪ Pancake	11.32%
4. Mung Dhal <b>Khichri</b>	▪ A combination of rice and legume	9.16%
5. Mung Spinach <b>Saag</b>	▪ A preparation of leafy vegetables	11.31%
6. Mung Amaranth Saag		11.24%
7. <b>Mungbean</b> Sprout Salad	▪ Green gram, <i>vigna radiate</i> var. radiata	10.66%
8. Sprouted Mungbean <b>Pulao</b>	▪ Rice cooked with vegetables	8.82%
9. Mung Sprout-Mint <b>Raita</b>	▪ Fermented curd with vegetables and species	7.18%
10. <b>Dahi Bhalla</b>	▪ Fermented and fried ball of mungbean in curd	9.55%
11. Mung Spinach <b>Pakoda</b>	▪ Fried snack prepared from vegetables coated with legume flour paste	9.73%
12. Mung <b>Namkeen</b>	▪ Fried and crispy snack prepared from legumes	10.42%
13. <b>Poha</b>	▪ Dish prepared from rice flakes and vegetables	10.70%

Data source: Bains, Yang and Sundar, 2003

Southern India. The results indicated that mungbean supplementation improved health parameters including clinical deficiency symptoms, body weight index, hemoglobin level and productivity (Vijayalakshmi et al., 2003). Hemoglobin level increased by 10% for school boys and girls receiving higher ID recipes (ID, 12%), while by 5% increase of hemoglobin level for the school children receiving

traditional mungbean recipe (ID, 7%). However, the one year feeding was insufficient to raise the hemoglobin level over the anemia level. The feeding trial demonstrated food-based approach on cost-effective plant based diet to improve iron deficiency, and suggested longer-term strategy to resolve severe anemia.

## Conclusions

These iron bioavailability studies, most of which employed in-vitro methodologies, produced principles and guidelines in household food preparations with vegetables to increase available iron in food. The findings were applied for recipe designs and conducting training of women's groups. The feeding trial has shown that the food-based approach using improved mungbean recipes cooked with tomato and other green leafy vegetables is a sustainable way to reduce nutritional disorder.

There are a number of factors to be considered in order to conduct a successful food-based nutrition improvement program. Besides a knowledge-base on nutrition and effective educational programs, a critical factor is the supply of recommended food items from the agricultural sector with a price within the economic capacity of the targeted population. An integrated program on nutrition and agricultural research is generally recommended. The sustainability of the nutrition program depends on the development of production technologies which can substantially improve the supply of needed food items. Research conducted at AVRDC has demonstrated that two technologies – high yielding mungbean and heat tolerant tomato – can be adopted in South Asia. The promotion of legume-based diet with tomato, green leafy and indigenous vegetables is to

raise dietary levels of protein, pro-vitamin A as well as available iron.

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