THE EFFECT OF COOKING METHOD UPON IRON AND ZINC BIOAVAILABILITY IN RICE.

By

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PLEASE NOTE

The greatest amount of care has been taken while scanning this thesis,

and the best possible result has been obtained.
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SUMMARY

Prevention of iron deficiency rather than treatment is preferred by most nutritionists. The presence of a maximal iron supply is not advised because an excessive or unnecessary iron supplement may have biological adverse effects such as impaired immune resistance or negative interaction with other trace elements.

All cereals and most vegetables contain phytate (inositol hexaphosphate) which can bind iron and zinc and reduce their biological availability. The amount of iron available for absorption in any given diet depends to a large extent on the composition of the diet, e.g. the presence of vitamin C and then haem iron, the absence of phytate. The complexing action of phytate appears to be an important etiological factor in the genesis of iron and zinc deficiency in regions of the world where the main staple food is rice. Rice is a staple food for a great proportion of the world population and the prevalence of iron deficiency is especially high in many countries including Iran, where there is high dietary rice intake. Rice is one of the most important staples in the Iranian diet and in fact rice for most Iranians is not a side dish but a main dish in which other foods or ingredients are put in rice, over rice or around rice. An awareness of cooking method effects on nutrient content is important because cooking techniques influence the overall quality of a group's diet.

The goal of this study is to determine the bioavailable iron and zinc in traditional Iranian rice dishes and to assess the effect of traditional cooking methods of rice upon iron and zinc bioavailability.
Iranians migrants residing in Sydney were surveyed in order to determine their food habits and traditional cooking methods for the assessment of their effects upon the bioavailability of iron and zinc in Iranian rice dishes. Approximately one hundred female Iranian migrants were surveyed by questionnaire with respect to their cooking procedures used for rice. Results indicate that the majority reported to consume rice either every day or at least 3 to 4 times a week (84%) and most soak their rice in salt water (50%) prior to cooking and that the most common cooking method is boiling (72%).

The bioavailability of iron and zinc for Sunbrown Natural Calrose (brown rice) and Basmati (white or polished rice) was determined. Rice contain phytate, which is a potent inhibitor of iron absorption and the amount of phytate in rice depends on whether the rice is polished or not. The pre cooking treatment such as soaking can also influence the phytate content of the rice. Soaking prior to cooking also reduced the amount of total iron and zinc. Basmati rice soaked in salt water had significant (P<0.05) increase in bioavailability (up to 32%). Reduction of phytate after soaking of rice for the period of 4 hours in salted water was most effective in increasing the bioavailable iron and zinc.

Unpolished brown rice which traditionally was used in ancient Iran is not very popular today due to the use of white rice. Although brown rice contains higher amounts of iron and zinc, the bioavailable iron and zinc was similar to white rice (0.23mg/100g). Using the traditional Iranian methods of cooking significantly (P<0.05) decreased phytate and increased the bioavailable iron (20.4%) in brown rice
after four hours of soaking in salt water. The bioavailable zinc (16.4%) as well as bioavailable iron (20.4%) was significantly (p<0.05) increased after 8 hours of soaking the brown rice in salt water.

Assessing the total and bioavailable iron and zinc after cooking shows that cooking via absorption method did not have any significant effect on the bioavailable iron and zinc in both Basmati and Sunbrow Natural Calrose whereas cooking via boil method significantly (P<0.05) decreased the amounts of total and bioavailable iron and zinc in Basmati rice.

The majority of Iranians reported using either vegetables or legumes in their rice dishes. The bioavailable iron and zinc were measured in different traditional rice dishes.

The inhibitory effect of phytate is also overcome by adding different herbs, vegetables and legumes used traditionally in the rice dish cooked by Iranian. Herbs or vegetables such as coriander, parsley and cauliflower used in Iranian rice dishes increased the bioavailable iron and zinc. Coriander significantly (P<0.05) increased the bioavailable iron and zinc whereas parsley increased the bioavailable zinc in traditional Iranian rice dishes.

Results obtained from this research show that method of cooking as well as the food mixture is very important for the absorption of iron and zinc from the diet.
DISCLAIMER

The author certifies that this report represents her own original work, that it contains no material which has already been published and that to the best of her knowledge, it contains no copy or paraphrase by another person or authority except where due acknowledgment has been made.

Signature

Mahnaz Shahnaseri
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CHAPTER 1

1. GENERAL INTRODUCTION

This thesis is an attempt to gain an understanding as to how and/or why certain food customs/or habits developed. In particular, it is looking at rice preparation methods and their optimisation for nutrient utilisation. These optimisations may be historical methods for avoiding nutritional deficiencies especially mineral deficiencies in the case of rice, the dietary staple of Iran.

1.1 Mineral deficiency

A nutritional deficiency of a mineral develops when either the amount present or absorbed from the diet is insufficient to meet normal requirements. Suboptimal mineral nutrition and especially iron deficiency is prevalent in both developed and developing countries. Iron deficiency is the most common deficiency in the world (Ramdath and Golden, 1989). Baynes and Bothwell (1990) proposed that iron deficiency due to inadequate diet is a major problem afflicting approximately 30% of the population in the world. Even in the industrialised western world iron deficiency affects at least 26% of the population (WHO, 1991). Hurrell et al., (1989) noted that there are several reports of iron and zinc deficiency in United States. Iron and zinc deficiency is especially high in populations consuming plant based diets due to poor
bioavailability of minerals in their foods caused by the effect of inhibitors such as phytate, phosphorus, tannins and fibre (Prasad, 1983).

Ramdath and Golden, (1989) maintained that a deficiency of iron may result in more problems than simply anaemia. The limitation of work performance in the presence of iron deficiency has been reported by several studies (Hamilton and Whitney, 1994; Eastwood, 1997). Beard et al., (1996) proposed that the negative effects of iron deficiency on work efficiency may have a huge economic impact. Lack of iron also can result in increased microbial growth and infection (Weinberg, 1984). The majority of chronic diseases are affected by mineral deficiencies (Goldberg, 1994). Iron is required for a number of important biochemical reactions in the brain and the function of the brain itself may be affected by iron deficiency (Youdim et al., 1980; Eastwood, 1997).

1.2 People at risk for mineral deficiency

Adolescent females are considered a nutritionally vulnerable segment of the population. A rapid growth rate combined with expanding blood volume and developing sexual maturation as well as poor dietary habits increases the risk of mineral deficiencies in this population (Pennington and Young, 1991). Several studies have reported a high occurrence of iron and zinc deficiency among teenage girls (Lee, 1978; Liebman et al., 1983). Iron deficiency is a common disease in the infant, teenagers and young women (Dallman et al., 1984).
Eating habits are an important part of lifestyle and often reflect an individual's philosophy (Whitney et al., 1995). Understanding the process of food choice is central to health promotion (Devine et al., 1998).

To identify the people at risk from mineral deficiency it is necessary to ascertain the food and nutrition knowledge and beliefs as well as to establish recent and remote food intake patterns. Anthropometry and selected laboratory investigations of specific foods may help in the identification of people at risk.

1.3 Food consumption patterns and minerals

Food consumption plays an important role in health. Hallberg and Rossander, (1984) proposed that the absorption of minerals in the diets of people in developing countries is often low due to a high concentration of factors inhibiting absorption.

In most parts of the world the average diet is predominantly cereal in nature, with wheat, rice and maize forming the major staple. Rice forms the second major cereal in human diets and is the most important source of cereal foods especially in Far Eastern countries. In many parts of Asia rice provides more than 70% of the energy in the diet (Garrow and James, 1993). Although rice is sufficient in minerals such as iron and zinc (Hallberg, 1974), its low bioavailability is one of the most significant factors for deficiency of these elements (Indumadhavi and Agte, 1992). Several studies suggest that iron and zinc absorption are low from rice based meals because metals in cereals are present as inorganic salts with low solubility (Agte et al., 1998). Due to the
high susceptibility of cereals for fat oxidation during storage, cereals are difficult to fortify with highly bioavailable minerals (Hurrel et al., 1989). Although rice contains many essential minerals such as iron and zinc, the bioavailability of these elements is only about 10% of the total (Mills et al. 1988). DeMaeyer and Adiels-Tegman (1985) reported that the prevalence of iron deficiency is especially high in many countries where there is high dietary rice intake. There is a high prevalence of iron and zinc deficiency in most developing countries (Hallberg and Rossander, 1984) which may have important negative effects on health and wellbeing. It is important to find a realistic, effective, cheap and acceptable method to counteract the iron and zinc deficiencies and improve the bioavailability of these minerals in the diet, by reducing the level of inhibitors and raising the level of enhancers. One of the major known enhancers of iron and zinc absorption is ascorbic acid (Skikne and Baynes, 1994). Ascorbic acid has the ability to chelate iron and zinc (Gillooly, et al., 1983). Therefore in a dish containing rice and vegetables, the amount of inhibitors such as phytate and enhancers such as ascorbic acid are the crucial factors in determining the bioavailability of the iron and zinc. Hallberg, (1981) proposed that the addition of cauliflower containing 70 mg ascorbic acid to a vegetarian meal trebled the amount of bioavailable iron. Different cultures with different cuisines use different vegetables in various ways. Ascorbic acid is only one of several organic acid constituents of vegetables and there is little or no information concerning the possible role of adding vegetables to rice dishes.
1.4 Iron and zinc in vegetables

Plants and/or vegetables provide a major part of the diet of most of the world's population, and are major sources of energy in most developing countries. Although vegetables are plentiful in minerals such as iron and zinc, minerals from plant foods are poorly absorbed in comparison to animal foods due to the presence of dietary fibre, phytate, oxalate, and phosphate which form insoluble complexes with non haem iron and limit its absorption (Eastwood, 1997). Thus the bioavailability of dietary iron in the diets in many developing countries is very low (Garrow and James, 1993).

Iron absorption may be significantly affected by the intestinal mucosa, total and chemical nature of iron in the food and other dietary factors that increase or decrease the bioavailability of iron (Herbert, 1987).

1.5 Food habits and nutritional anthropology

The study of the social life of human populations is called social anthropology, while the study of customs in different societies is termed ethnology. Nutritional anthropology is the study of the foodways or eating pattern of men of the past and present. Murcott (1988) believes that social anthropology has paid a great deal of attention to the study of food habits. She believes that social anthropology and sociology are closely related and each is less easily classified as a unitary discipline but more as a plural basic which consist of a number of theoretical perspectives. This
means that any aspect of human activity has to be studied in relation to the social context in which it occurs. Food habits research is an excellent example where the basic social environment of food production and consumption is experienced (Germov and Williams, 1999). In other words, to understand food habits social patterns of food consumption must be taken into account.

Stanton (1986) stated that the favourite foods are those foods which people learn to like early in life. People in various countries have adopted traditional eating patterns which are mostly associated with the food available. Choice of foods or food habits is not only affected by culture, it is under the influence of a variety of factors such as:

- Social, (Wein et al., 1989)
- Religious, religious beliefs can strongly govern food selection (Stanton, 1986),
- Personal (taste habits), eating habits are based on personal beliefs as well as the taste habits developed mainly in childhood (Stanton, 1986),
- Nutritional knowledge of individuals, although people need nutrients, they do not eat nutrients, they eat food (Sims, 1978),
- Immigration, (Hopkin et al., 1980; Manderson, 1986), Migration affects individuals in many ways, it often causes change in immigrants’ lifestyle and attitudes, one of the most important changes is in the food consumed as well as in dietary patterns (Shahnaseri and Bergan, 1994),
- Generation changes (Freedman and Grivetti, 1984). Although food habits has been defined as a “standardised set of behaviour” by Mead, (1943) Nalbandian
(1981) stated that food habits are affected by alteration in social and/or cultural environments,

- Income (Dennis and Shifflett, 1985),

- Mothers beliefs and interest (Burton and Foster, 1988),

- Acquired tastes (Germov and Williams, 1999).

In general, all the above factors would influence the nutritional status as well as health of the individual. Stanton (1986) proposed that some of the health problem of migrants in the Australian community were related to food due to dietary changes in which total food increased while real needs decrease. Migrants reduced their starch intake while sugar, fat and meat consumption increased.
1.6 Objectives

In general the aim of this study is to investigate iron and zinc bioavailability in rice from traditional ethnic cooking methods. Very little information currently exists on rice pre-treatment and cooking methods as used by different ethnicities.

Specifically the objectives of this study are:

1- To determine the traditional methods of precooking and cooking of rice by Iranians.

2- To ascertain the effects of preparation and cooking methods of rice on:

- Total iron and zinc present
- Bioavailability of iron and zinc

3- To determine the effect of additional selected traditional herbs, vegetables and legumes upon iron and zinc bioavailability.
1.7 Significance of the study

This research will provide information about methods of precooking and cooking of rice as well as determining the available iron and zinc in various rice dishes. Once this awareness has been established these cooking methods and food behaviours can be used to understand the development of food habits. In addition this information can be used to educate and increase the awareness of all rice consumers in developing countries particularly where access to meat, poultry and dairy products is minimal. It is also useful for vegetarians.
CHAPTER 2

2.1 LITERATURE REVIEW

2.1.1 Dietary patterns and iron and zinc

Food habits are acquired early in life and once established are likely to be long lasting and resistant to change (Fieldhouse, 1995). What people eat and their nutritional status is strongly influenced by their food habits and eating patterns. The iron and zinc content of the diet and its bioavailability are reflected in iron and zinc status of the people. A high incidence of iron deficiency is seen in developing countries which is often due to the limited content of food enhancing the absorption of the non-haem iron (Hallberg and Rossander, 1984). Various socio economic, cultural, ecological, educational and personal factors can affect food habits and eating patterns (Djazayery and Kholdi, 1992). The method of preparation and the presence of enhancers and or inhibitors may also effect the bioavailability of minerals such as iron and zinc (Acosta et al., 1984). Therefore the type of diet consumed by individuals has a significant and independent influence on iron status that modifies the effect of iron requirements. A well selected diet of high iron bioavailability plays a role considerably more important than does iron supplementation in combating iron deficiency (Monsen et al., 1983).
2.1.2 What is cuisine?

Cuisine is a term used to define a style of cooking, preparation methods and techniques of eating of normal or typical food of a particular country. Cultural groups throughout the world have developed characteristic cuisines. Farb and Armelagos, (1980) proposed that the origin of most cuisine is lost in the unrecorded past. These cuisines are based on foods that are readily available in the local area and therefore markedly differ from one area to another. Often foods prepared and eaten in one region do not feature in the cuisine of another region. Individuals who have grown up with these foods accept them as part of their cuisine although strangers may regard them with suspicion (Burnett-Fell, 1994). Rozin (1983) proposed that each ethnic group fashions its food stuffs somewhat differently from the next, yet the general behaviour of cooking always seems to form certain patterns, and to follow rules that can be demonstrated.

2.1.3 Culture and food

Culture in its more anthropological sense refers to the systems of signs, meanings and world views of a particular group of human beings. MacLachlan (1997) proposed that “If we are to avoid being churned in a mono cultural melting pot then this requires us all to acknowledge, tolerate and work with different interpretations of some of the things which we hold most precious.” One of these things is food pattern. Germov and Williams (1999) identified food as a factor to
build and maintain social relationship in all cultures. The consumption of food is a social occasion because in most traditional cultures the family members and friends gather together for the purpose of eating. The process of growing, harvesting, processing and preparing food mostly involves the efforts of multiple individuals working together (Rozin, 1996). Therefore, food is an extremely valuable social instrument for people. The offering or acceptance of food by one person or group to another is generally viewed as a sign of friendship and a willingness to strengthen or establish a friendship and family relationships (Germov and Williams, 1999). Food is shared with a friend but not with an enemy (Foster and Anderson, 1978). Bass et al., (1979); Farb and Armelagos, (1980) proposed that food does not merely satisfy hunger and nourish the body, but it also is used to initiate and maintain relationships or friendship, as well as determining and demonstrating the extent and nature of the relationship.

Food is often used to share happiness as well as being a welcome gift for new neighbours. Food is not only essential for survival, it is also one of the great pleasures of life. Many social occasions are organised around food. The interplay between culture and food is complex. It is now very easy to travel anywhere in the world and observe the signs of cultural difference where commodities such as restaurant meals or items of clothing draw our attention to their ethnic origin. Casimir (1991) stated that culture does influence diet to a great extent. Hence culture is a major element that determines what kind of food, is consumed by each community and it is a life long process that makes groups of people similar to others. In addition, geographical, social, nutritional, economical and religious factors help form the nature of food intake. Human food habits may be described by the food
chosen, the method of preparation, the eating technique and the time and number of meals served during the day as well as the size of portions consumed.

Germov and Williams (1999) proposed that in western culture, nutritionists view food as a source of nutrients where as in some cultures such as Chinese or Vietnamese, food selection is not based on the physical needs of the body. These cultures consider wellness to be harmony and balance in body, mind and spirit (Bodeker, 1996). Lowenberg et al., (1974) also proposed that food is necessary for the ego as well as the body. According to Giff et al., (1972) for most people the symbolic meaning of food is more important than rational meanings.

For a person during times of stress food has different meanings; for example during examination time students eat more or sometime less than usual, and specially they eat high energy snack foods which were not included in their normal eating pattern (Bass et al., 1979). According to Giff et al., (1972) teenagers sometime refuse to eat foods they accepted earlier in order to show their independence. Boss et al. (1979) proposed that the interaction between some personal characteristics and culture determine an individual’s food behaviour.

2.1.4 Culture and cooking food

“Cooking is a universal and distinctively human activity”. Regardless of the way of choosing food, all people transform most of what they eat by heating them
through different fashions. Human beings are the only creatures who do not eat their food in the state that they capture it and unlike most animals not only wash and peel their food, they even preserve it by drying, cooling or smoking as well as cook it. Murcott (1988) believes that the human being has a dual character. Man is a species of animal more or less like other animals with parallel bodily functions and similar biological requirements for survival. On the other hand man is not an animal but human because he has language, culture and intelligence. Thus, Murcott (1988) defines humans as creatures of nature and culture.

Levi-Strauss (1970) called cooking and language as two human universals which are linked and similar. Therefore, analysing the structure of language could be used to reveal the structure of cooking activity and they both are a form of essential human expression.

The nutritional value and safety of many foods can be altered by how they are grown, processed and cooked. These changes are important considerations when food is selected. Although cooking and processing can alter the nutrient composition of foods, Mc Nutt and Mc Nutt (1978) proposed that the effects of processing upon the nutritive value of food is mostly considered by a food technologist rather than a nutritionist. Alteration of the nutrient composition of foods does not affect all nutrients of all foods. The most important nutrient losses due to processing and cooking are minerals and vitamins.
2.1.5 Foreign foods

Western diet no matter how much people enjoy it, is far from perfect nutritionally (Sprientsma, 1999) with many western diets having problems such as:

- Low nutrient density,
- High in sugar and fat and therefore provide more energy than we need,
- Low in fruits and vegetables,
- Neglect of dairy products.

(McNutt and McNutt, 1978)

Awareness of the dietary patterns of other populations offers several advantages that are more important than simply being cultural curiosities. Correcting the nutritional problems of a diet would be easier if greater awareness of other cultures food patterns was evident. McNutt and McNutt, (1978) proposed that some ethnic diets selected by other cultures have nutritional merit. They also mentioned that the selection or avoidance of any food could alter the nutrient composition of a diet. Thus, if the good points of other cultures food habits can be identified, successful replacement of most poor diet habits can occur.
2.1.6 Food and public health

The most powerful determinants of the health of populations are the cultural circumstances in which people live as well as their social and economic status (Blane et al., 1996). To promote the health of a population, social, economic and cultural circumstances in which health is created needs to be considered. The health promotionist particularly focuses on changing life style behaviours, including dietary behaviour in order to prevent disease in individuals (Tesh, 1988; Richmond, 1998).

Diet can be a part of both disease therapy and disease prevention. MacLachlan (1997) mentioned the importance of preventing the conditions which cause disease. He also proposed that psychological factors in the form of an individual’s health related behaviours such as sleeping or eating habits can have a dramatic effect on mortality. Germov and Williams (1999) stated that more attention is paid to the sociological study of food and nutrition than to the recognition of cultural differences that those cultural differences play in food consumption as well as the social implications of food upon health.

2.1.7 Food habits, why people eat what they eat

Food is the most basic necessity of life for all creatures in the world. Food habits are dependent on attitudes and general culture which are acquired early in life. They are an integral part of the life style and form a pattern of behaviour which is
characteristic of a group (Davidson, et al., 1986). It is learning what is expected of
one within one's own culture under particular circumstances. According to Stanton
(1986) people choose foods due to availability and price. Meade (1997) claimed
that usually people with a lower socioeconomic status in Australia choose high fat
and low nutritional value foods such as McDonald's, Kentucky Fried Chicken, hot
chips and pizzas. Wealthier and better-educated people may make better choices by
purchasing more nutritious foods. In comparison the average amount of money
spent by low income people (lowest 20 percent) on meals eaten out of the home is
one third of the money spent by the highest 20 percent (ABS, 1996). The
distinctions within groups are cultural and also political (Lamont and Fournier,
vary in different parts of the world, there are still similarities in the way people of
different cultures think about food.

Bass et al., (1979) proposed that the foods which are preferred by people are
not necessarily consumed. Preference for a food is just one of the factors involved
in choosing food. Culture, social background, tradition, religion as well as taste
habits which developed mainly in childhood are all important factors for choosing
foods (Djazayery and Kholdi, 1992). The variability of the cultural system causes
variability in human patterns of food selection (Thomson, 1988). Advertising also
influences our food choices. Nutritious foods are not always chosen due to
ignorance (Stanton, 1986).
2.1.8 History of rice production

Cultivated rice is designated as either *Oryza sativa* or *Oryza glaberrima*. *Oryza sativa* is the predominant species where as *Oryza glaberrima* is grown only in Africa on a limited scale (Marshal and Wadsworth, 1994). The true rice (*Oryza sativa*) has “reigned supreme” in South and East Asia for 5000 years. According to Chang (1976) rice was cultivated between 200 and 1500 B.C. in an area extending from central India, through northern Burma, northern Thailand, Laos, Vietnam and into south eastern China, then spread to Indonesia, the Philippines and northern Australia. Persian rice was first cultivated in the Caspian area around the fourth century B.C. The best Persian rice still comes from Gilan by the Caspian, where it is also the staple diet, eaten for breakfast, lunch and dinner (Khalili-Batmanglij, 1992).

Rice is a plant grown in hot climates in which the evaporation of water must be taken into account, because the main shoots cannot tolerate dry conditions for more than a few days and the quality of the crop depends on the amount of water present. Water and warm temperatures stimulate rice to grow fast and with high yields. Rice can be grown under a broad range of climatic conditions as long as the essential requirement of water is present. There are eight thousand varieties of rice which can be classified into three kinds depending upon the length of the grain. Traditionally, rice varieties are classified as long, medium and short grain types (Toussaint-Samat, 1993). Rice growing requires a large labourforce, and according to Toussaint-Samat (1993), many nutritionists believe that the production and
consumption of rice as a staple food may influence the character and behaviour of the people.

2.1.9 Production and consumption of rice

Rice is the main staple food for about three quarters of humanity. Approximately 90% of the world’s rice is produced and consumed in Asia. According to Marshal and Wadsworth (1994) the overall increase in production and consumption of rice in recent years signifies the value of rice as a food.

Although China followed by India are the first and second rice producers in the world, due to their large populations which consumes almost all of the rice production, Thailand and United States are the two top rice exporting countries (Marshal and Wadsworth, 1994). The consumption of rice in Asian countries where rice is grown is 100 to 200 kilo gram per person per year (Toussaint-Samat, 1993). The consumption of rice in the United States doubled since 1975 and recently was 10 kg per capita (USDA, 1992) and in Australia Rice intake increased 11.7% to 7.3 kg per person (ABS, 1998).

2.1.10 Nutritional value of rice

Rice without hulls contains about 80% starch, 8% protein and 12% water. Due to the fact that rice is such a predominant food in the diets of some nations it’s nutrient composition is relatively as important as the nutritional composition of
wheat in western countries. Ensiminger et al (1994) stated that although rice has a good nutritional value in comparison with the other major cereals used as food staples around the world, people in most of the rice consuming areas of the world have a high incidence of disease resulting from insufficient and improper diet.

In the rice eating nations approximately 60 to 80% of the energy comes from rice. Therefore only 20 to 40% of the diet includes vegetables, meats, fish, fruits and other foods which are needed to make up the lack of nutrients such as vitamins and minerals (Ensiminger et al., 1994).

2.1.11 Traditional rice preparation

In some cultures, rice cooking is as simple as putting rice in a pot of water and boiling until tender. In other cultures, especially those where rice is the staple food and the main ingredient of the meal, rice preparation is a more serious affair. This can probably be best illustrated by the rice preparation practice in countries such as Iran. In Iran much time and attention are devoted to the preparation of rice before cooking. The principal step in this preparation is the washing phase. Ordinarily it may seem that washing rice is not really necessary, and it is often recommended that rice not be washed in order to retain the required enrichment. However, in Iran or other rice centred cultures such as in Japan, washing is an unavoidable part for the proper preparation of rice. Washing rice in the Iranian culture is usually more than just a simple rinsing. It is a process of adding water,
scrubbing the rice by hand, pouring off the water, and adding more water again and
again until the water finally runs clear. Marshal and Wadsworth, (1994) proposed
that the Japanese wash their rice for better taste. Even in well milled rice, some bran
residue usually adheres to the kernel surface and depending on the age of the milled
rice the residual bran can oxidise and produce free fatty acids that can be detected as
an off taste or rancid flavour.

2.1.12 Food history of migrants

2.1.12.1 Asian diet

The traditional diets of Asia, including China, Japan, Korea, India, Thailand,
Vietnam, Cambodia, Indonesia and other Pacific rim countries could be responsible
for low rates of disease. According to Claiborne and Timothy Ryan, (1997) most of
the daily energy of Asians are derived from foods such as rice, noodle, millet, corn
and other grain. Vegetable oil rather than olive oil is the preferred cooking fat. Fish
and shellfish are served less frequently. Egg, poultry and sweets are not included in
a typical daily menu, although they may be included once or twice a week. Red
meat occupies the very top of the dietary pyramid in the Asian diet (Claiborne and
Ryan, 1997).
2.1.12.2 Iranian diet

Iran, historically shows strong common and distinctive themes in its culinary cultures, particularly in the Persian-speaking centre: distinctive and elaborate rice cookery, specific varieties of kebab, fruit and meat, sweet and sour, sweet and savoury combinations, distinctive spices, especially the use of fenugreek and tarragon. Of the non Persian speaking regions, Azarbeyjan and particularly, Tabriz enjoy a high reputation for food. Distinctive foods include the unique Kofteh Tabrizi, a large parcel whose outer cover consists of a paste of meat and rice stuffed with chicken, prunes and eggs. The Caspian provinces feature special styles in fish cookery, in turn quite distinct from the fish cookery of the Golf province of South of Iran (Zubaida and Tapper, 1994). Seasonal fruits are usually served after tea. Tea is served in “see through” glasses (never with milk). Iranians are very fond of fruit and when fruits are in season they eat them on any possible occasion. Iranians serve fruit at lunch, tea, or dinner, often as a bowl of cold fresh fruits. Fruits include cherries, peaches, strawberries, grapes, melons, oranges, mandarins and cucumbers. Iranian cucumbers are small with a delicate flavour and are served as a fruit.

Traditionally Iranians drink cold water with their meals. Every province of Iran has its own speciality for making sweets, biscuits and candies. Sweets made of dates, rice and many other fruits and substances are very common in Iran.
2.1.12.2.1 Rice cookery in Iran

Although all provinces of Iran have their own dishes and specialities, the national dish in Iran is rice prepared in several special ways and served in large helpings with almost every main dish. In fact, very few main dishes would be considered complete without it (EIRI, 1998). Iran features an elaborate culture of rice cookery which seems to go back in history to at least the Safavid times in the 16th century. There are two types of rice dishes in the Iranian diet, chelo and “polow”. Typically, chelo is plain boiled and steamed rice which is then combined with various stews or ragouts. Polow is rice with tasty ingredients mixed with the boiled rice before the second part of the procedure (steaming). Both kinds of preparation require two steps, first boiling and then steaming (Khalili-Batmanglij, 1992).

Chelo is rice prepared in several stages for up to 24 hours, boiled and steamed and served separately with all type of meat dishes, while polow is rice cooked with other ingredients. Iranians prefer rice which is fluffy and tender, not sticky and soggy. Saffron is frequently used to flavour and colour rice.

2.1.12.2.2 Change in Iranian polow preparation

The art of preparing Iranian polow developed from the end of the 16th century to the present day. In a 70 to 80 year period during the 16th century about 70 different recipes for preparing polow were invented. During the 19th century another
cookbook written by a royal cook from Qajar Tehran, presented 29 recipes for
polow. About 30 percent of these 29 recipes were already found in the recipe
collections of Abbas the Great’s cook. About 60 percent of recipes were forgotten
between 1600 and 1850. They obviously fell out of use when they lost their
attractiveness in terms of fashion in the course of history and the rest remained as a
limited range of dishes belonging to what gradually became classical Iranian cuisine.
According to Zubaida and Tapper (1994) a recent Iranian cook book contains
twelve to fourteen recipes roughly presenting the range of polow recipes practised in
an average Iranian middle class household.

2.1.12.2.3 Origins of Iranian rice traditions

"The Persian word for rice berenj comes from the Sanskrit vrihi and was
probably brought to Iran from South-East Asia or the Indian subcontinent and it was
cultivated in North of Iran around fourth century" (Khalili-Batmanglij, 1992). In the
period from the Arab invasion of the Iranian plateau until at least Seljuk rule, rice
was not at all common among the Iranian peoples. Eastwood (1997) stated that
during 13th century rice was a luxury in Persian cuisine and was mixed with meat
into a “Pilau style” dish. Today rice is eaten in great quantities (breakfast, lunch and
dinner) in Caspian region, but elsewhere in Iran rice is eaten in their main meal
(lunch). Rice is considered a luxury by the peasants or the urban poor and eaten by
them only on special occasions or served to guests (Khalili-Batmanglij, 1992).
There are five major varieties of rice in Iran. Today imported Basmati rice is readily available in Iran and Australia and it is similar to Persian rice.

2.1.12.3 Australian diet

The Aborigines populated Australia for at least fifty thousand years before the first white settlements were established in Australia. The Aborigines were found all over Australia and were known as a hunter-gatherer society. Aboriginal diets were varied depending upon where they lived and therefore their food and eating was regionalised (Riddle, 1989). The Aborigines consumed both animal and plant foods and they had a distinct lack of fat in their diet (Argyriou and Mallos, 1988). The current Australian food habits basically started in 1788 with the first settlement led by Captain Cook. The first fleets carried on board flour, rice, salted meat, sugar, salt, livestock, alcohol, vine cuttings and seeds (Riddell, 1989).

In the nineteenth century, Australians “perpetuated” an English style of cooking with English food, structure and values. During this time due to the abundance of mutton in Australia, meat became the major component of the diet (Santich, 1995). Later the settlers from other countries such as China, Italy, Germany and Greece came to Australia. They introduced their own food habits to Australia. A major influence on Australian’s food habits is due to immigration. It
took years before these immigrants food habits were accepted. At first when a
different culture settled in Australia, due to the language problem, their food habits
remained isolated (Riddle, 1989).

In the early twentieth century, with discovery of vitamins, Australians started
to realise the benefits that nutrition had to offer. It was during this time that the first
promotions on Australian eating were conducted. It was also during this time that
many European influences on the Australian diet were adapted. Some migrants
opened up restaurants that featured their traditional cuisines. One hundred years
after the Chinese immigrated to Australia their foods began to be accepted (Riddle,
1989).

According to Chapman et al. (1998) Australians are moving towards a more
varied and adaptable cuisine which reflects the nature of its climate and diverse
population. Foods such as prawns and lobster are increasingly preferred. Even
Christmas fare is likely to be influenced by the weather with salads, ham and poultry
being served instead of the traditional English Christmas dinner.

During the twentieth century processing technology has given Australians
microwave ovens as well as freeze-dried foods. Lifestyles have changed too. From
the mid 1970’s due to the time demands of working women convenience foods have
entered the market. Working women brought a significant change in food habits
with more and more convenience foods as well as take away foods being consumed
(Santich, 1995).
Ross (1995) proposed that native fruit, nuts, berries, fish and meat may help Australian to create a unique cuisine. Australia has some unique and very unusual foods such as vegemite, lamingtons, damper, Anzac biscuits, iced Vovos, curried yabbies and rice (Holden, 2000).

Certain geographical locations throughout the country reflect the ethnic population within that area, in turn influencing the local cuisine. For example people who live in the Barossa valley have a unique blend of German and English cuisines (Barossa, 1999).

2.1.13 Iron biochemistry

To interpret the behaviour of iron in foods some understanding of the chemistry of iron is necessary. Iron (atomic weight 55.85, atomic number 26) is the second most common element on earth (Ziegler and Filer, 1996). Depending on the chemical environment, iron itself may occur in foods in several different forms, such as elemental iron (Fe⁰), and in insoluble and soluble forms. Elemental iron (Fe⁰) is rarely found in biological environments but does occur in food as a common food additive. The soluble form may be in complexes or ionic. In aqueous solution, iron exists in two oxidation states: the ferric form (FeIII) and the reduced ferrous (FeII) form. These two forms of iron are the only states which are stable in an aqueous
environment. Thus they are the only states which occur naturally in foods. Iron is able to easily change between the two forms (Fe II and Fe III) which enables iron to serve as a catalyst in redox reactions by accepting or donating electrons (Lee and Clydesdale, 1980; Ramdath and Golden, 1989). In the presence of oxygen, Fe (II) is rapidly oxidised to Fe (III). Fe (III) is extremely insoluble in water, therefore it precipitates as oxides of iron. "The solubility of ferric oxide is so low that at neutral conditions there are only a thousand iron atoms per cubic centimetre of water, far too little to be of any practical value to a living cell" (Emery, 1982).

Iron is an essential nutrient for most living organisms. It is an essential cofactor for hundreds of enzymes and is used in protein metabolism (Beard et al., 1996). It is found in all human cells. Body iron can be considered as being in two main components: functional and storage iron. Iron is essential for the formation of haemoglobin the red pigment in blood, myoglobin and iron dependent enzymes (NRC, 1989). Iron in haemoglobin combines with oxygen and transports it through the blood to the body’s tissues and organs. The body contains between 3.5 and 4.5 g of iron, 2/3 of which is present in haemoglobin. The remainder is stored in the liver, spleen and bone marrow. A small amount (about 5%) is present as myoglobin, an iron protein complex, which acts as an oxygen store in muscle tissue.

Iron is also present in many enzymes in which it is a cofactor involved in the production of energy. Iron is important in maintaining many of the healthy functions of the immune system. Also, some iron is used in the formation of other cells and is
transferred to and from different storage compartments but the main part of the internal iron metabolism is a recycling of iron in the red cell mass. Brabin and Brabin, 1992 stated that iron might even be essential for skeletal growth. Although most iron problems are due to a lack of iron, either too much or too little iron may create problems (Lieberman and Bruning, 1990).

Despite the fact that iron is the second most abundant mineral in the earth's crust, and also the ingenious mechanisms present in the body to maintain iron balance, the majority of nutritionists believe that iron deficiency is a serious health issue in many parts of the world (Hallberg, 1982; Beard et al., 1996).

Iron deficiency will occur when the dietary intake does not meet the body's demands. The main causes of iron deficiency which are due to inadequate absorption or increased loss are listed as follow (Andrews, 1999):

- Poor bioavailability
- Antacid therapy or high gastric pH,
- Excess dietary phytate,
- Gastrointestinal blood loss
- Parasitosis
- Chronic infection
- Plumonary blood loss
- Infection
2.1.14 Dietary source and absorption of iron

The amount of minerals varies between foods. Iron occurs in two forms in foods (haem and nonhaem). Haem iron is broken down within mucosal cells by haem splitting enzyme (Hallberg, 1982). The absorption of haem iron is up to ten times more than others. The average absorption of haem iron by healthy people is about 23 percent. Some foods have much more available iron than others. Deficiency of iron is relatively insignificant among people who consume greater numbers of servings of the animal food groups. The most important source of iron are those foods which are eaten frequently, those which are rich in iron and those from which iron is well absorbed. Liver, kidney, lean red meat and poultry all contain iron which is known as haem iron or organic iron (Inge, 1993) which is the most absorbable (10-30 percent). Although liver is one of the best sources of iron, it is not recommended for management of iron deficiency due to its high cholesterol and potentially high carcinogen content (Krummel and Kris-Etherton, 1996). Red meat is a major source of iron. Other substantial sources may be eggs, cereals and green leafy vegetables.

Plants contain nonhaem or inorganic iron. Hallberg and Rossander-Hulten (1991) noted that nonhaem iron contained in cereals and pulses may be a main source of dietary iron especially in developing countries. Hallberg (1974) proposed that nonhaem iron is the main source of iron in the diet, because the amount of haem iron in the diets of people in developing countries is negligible and also inspite of the
high meat consumption of western type diet, haem iron contributes only 1-2 mg per day. Boyle and Zula (1996) proposed iron and zinc as two minerals that should be of concern for all vegetarians. Absorption of non-haem iron is highly variable between 2 to 20%. This absorption depends on three main factors: dietary iron content, the nature of the meal and the iron status of the individual or the amount of storage iron.

2.1.15 Dietary iron contents

Reilly (1996) stated that the percentage absorption of iron as well as zinc decreases when the intakes of these minerals are high. Hallberg (1982) stated that the dietary iron absorption in subjects with iron stores of 500 mg was reduced by half. Also a rapid decline in iron absorption occurs in the case of daily administration of relatively high doses of iron (Norrby, 1974). Therefore, Liu et al. (1995) recommended supplementation with weekly doses of iron to be considered among iron deficient groups.
2.1.16 Nature of the food

Determining the amount of bioavailable iron or iron that is physiologically useful to the body rather than total iron in foods is well recognised and supported by several studies (Fairweather-Tait, 1987). The bioavailability of iron varies markedly between diets of different populations. Hallberg and Rossander (1991) proposed that the composition of the diet may have a several fold influence on the absorption of minerals from foods. There are a few foods which affect iron absorption. They contain chemical compounds that can interfere with the body's ability to use iron. Cereals and many high fibre foods contain phytate, a substance that several studies have reported to trap or bind iron and make it un-absorbable. The absorption of non-haem iron is determined by the composite effect of several factors promoting or preventing iron absorption. Depending on these factors, absorption of iron from a meal may range from 1 up to 40 percent in the same individual (WHO, 1988).

The British Nutrition Foundation (1995) reported that cereals provide approximately half the average household intake of iron. The low bioavailability of iron in cereals is also reported by several studies (Indumadhavi, 1992; Tuntawiroon et al., 1990).

Evaluations of diet for iron adequacy require knowledge of both the amount and the bioavailability of the iron in the foods. In the aetiology of iron deficiency, low iron availability is considered to be one of the most significant factors (Brian et
Rao and Parbhavati (1978) stated that low availability of dietary iron is one of the most important factors which leads to iron deficiency. Therefore for evaluating iron deficiency it is very important to consider the availability of dietary iron (Schricker, et al., 1981). The intake of iron may be low in a western diet due to its high sugar and fat content (Whitney et al., 1995).

2.1.17 Iron status of the subject

The absorption of iron is influenced by the iron status of the subject. More iron is absorbed by the iron deficient and less by the iron replete individual (Hallberg and Rossander (1991). Garrow and James, (1993) stated that the absorption of iron may double in the case of iron deficient subjects with no iron stores. Cook et al., (1991) demonstrated that the iron stores of an individual are the principal determinant of iron absorption. To meet excessive iron demands as in late pregnancy, the body can use the specific storage protein, ferritin, which stores iron in the body (Hallberg, 1982). Hallberg (1981) proposed that the bioavailability of iron should be expressed as an absorption value. The body also has the mechanism to regulate the absorption of iron when it is affected by actual requirements (Hallberg, 1982). Cook (1993) stated that the body increases the rate of iron absorption when there is an increase iron loss in the case of bleeding or menstruation and a decreased iron absorption in iron overload. Hallberg (1982) stated that iron status variation may lead to over a 10 times variation in absorption. Iron status of
individuals controls the absorption of iron as well as the distribution and transport of iron to different tissues (Hulten et al., 1995).

2.1.18 Zinc biochemistry

Zinc with atomic no. of 30 and atomic weight of 65.38 is a small ion (0.065nm). The essentially of zinc has been known for more than 100 years (Garrow and James 1993). Zinc is a key nutrient for optimal functions in the body and is present in the tissues and fluids of the body. More than 95% of the body zinc content is intracellular (Ziegler and Filer, 1996). The body contains between 2 and 3g of zinc. Zinc is a trace metal nutrient that influences many physiological functions, including immunity. It is a constituent of over 80 enzymes, (Liberman and Bruning, 1990). Zinc is associated with the hormone insulin, which helps regulate the body's blood glucose level. Garrow and James (1993) stated that a lack of scientific basis exists for setting recommendations for dietary intake of zinc. However zinc must be replenished daily (Boyle and Zula 1996).
2.1.19 Dietary source and absorption of zinc

Foods greatly vary in their zinc content. Cereals are the major source of zinc in large parts of the world. A cereal based unrefined diet may contain a large content of zinc (Garrow and James 1993). The amount of total zinc from cereals is dependent on the degree of refinement of the grain, because zinc is mainly located in the outer layer of the grain and the majority of the zinc content may be removed through processes such as milling. Several studies reported that foods of vegetable origin are usually low in zinc (Agte et al., 1997).

The content of zinc in vegetables is dependent on variety, class and growing location. Davis et al. (1984) reported up to 10 fold variation in the zinc content of cereals. Although zinc composition is dependant on the availability of zinc in the soil to the plants, different varieties of cereal apparently absorb different levels from the same soil and the same variety may absorb different amounts of minerals from different soils. The bioavailable zinc is the amount of zinc which is absorbed and used. Zinc bioavailability is approximately 5 to 10% from cereals based diets. Total zinc content of the diet provides only a gross estimate of zinc intake. Several components in the foods provide ligands for zinc binding; some improve zinc absorption whereas some inhibit. The inhibitory factors in zinc absorption are phytate, oxalate, fibre and tannin (Mills et al., 1988) while factors such as ascorbic acid increase zinc absorption (Agte et al., 1997). Zinc is most likely released from the food matrix during digestion and associated with low-molecular weight ligands.
such as amino acids, organic acids and phosphate. The degree of zinc absorption strongly depends on those low molecular weight substances. The absorption of zinc in or out of food can vary from 5 to 80% (Sandstrom and Lonnerdal, 1989). Sandstrom and Lonnerdal (1989) also stated that inorganic iron in pharmacological doses could decrease zinc uptake. Although zinc is most abundant in foods of high protein content, such as meat and nuts, the presence of other trace elements, inhibitors and fibre in foods may adversely affect zinc absorption (Whitney et al., 1995).

2.1.20 Iron and zinc deficiency

Iron deficiency (anaemia) is an important nutritional problem in the world, and is still common not only in underdeveloped but also in developed countries (Charoenlarp et al., 1988; Lehmann et al., 1992; Yip, 1994). Stoltzfus et al. (1997) claims that iron deficiency should be a global priority in public health, because iron deficiency affects more people in the world than any other form of malnutrition. Most nutritionists believe that the most frequent cause of iron deficiency is nutritional (Hallberg, 1982). The principal reason is not a low dietary intake of iron but rather a low bioavailability of iron present in the diet.
Nutritional iron deficiency usually develops slowly over months or even years and it is associated with periods of heightened physiological requirements. The earliest phase of iron deficiency is accompanied by a gradual depletion of storage iron (Cook, 1990). Whitney et al., (1995) proposed that iron deficiency begins when the absorption of iron can not compensate for losses or low dietary intakes and also stores are depleted. The term, iron deficiency, refers to the depletion of iron in the red blood cells and results in a lower haemoglobin concentration. In anaemia new red blood cells are smaller and lighter than normal. The most severe form of iron deficiency represents anaemia. The distinction between iron deficiency and anaemia is important. Therefore not all individuals with iron deficiency have evidence of anaemia (Dallman et al., 1984).

Prasad (1978) proposed that deficiency of iron had been known to occur concomitantly with zinc deficiency in many parts of the world. The first reports of zinc deficiency and growth failure in adolescents in Egypt and rural Iran occurred in the early 1970s (Halsted et al., 1972). Zinc and iron deficiency is more prevalent in countries that consume cereal and plant based diets. Thus their diet is low in animal products and high in phytate and fibre (Prasad, 1983: Davies, 1988).

The absorption of zinc is very low in modern societies due to a typical refined diet with a high fat content. It is also low in other parts of the world due to the presence of complexing agent such as phytate (Halstead et al., 1972).
2.1.21 Recommended dietary intake (RDI) for iron and zinc

Minerals are nutrients that are essential to life and belong to two groups: the macro and the micro or trace minerals. Iron and zinc are both classified as trace minerals and are needed in relatively small quantities (Liberman and Bruning, 1990). The micronutrients requirements are usually dependant on the amount required to support the equilibrium of the amount required to compensate for endogenous loss as well as the amount needed to maintain normal function.

2.1.21.1 Iron

Iron needs vary depending on age and sex. In an average day, an adult male needs to replace approximately 1 milligram of iron. The requirement of iron increases during puberty and in the case of growth and weight gain (Sjolin, 1981). Women in their childbearing years need to replace about 1.5 milligrams per day. Garrow and James (1993) proposed that two main facts (the iron status of the body and the bioavailability of iron in different diets) must be considered to translate the absorbed iron requirements into dietary iron requirements. Whitney et al. (1995) considered iron as a nutrient of special concern.
In young males from the age of 12 years, two factors affect the iron requirements. These factors are: peak growth rate which is approximately 1 kg per year higher than that in girls (Dallman et al., 1980) and the rate of expansion of the blood volume as well as body mass which is usually higher in young males (Truswell, 1990). Most nutritionists believe that iron needs of teenage females increase as they start to menstruate. Hallberg and Rossander (1991) recommended an iron intake of 19 mg per day in order to meet the iron needs for 95% of teenage females. Pregnant women and those on restricted diets need an additional 15 milligram daily for "expanded red cell mass in the mother and iron deposition in the products of conception" (Feltman, 1993; Ridwan, et al., 1996).

2.1.21.2 Zinc

Zinc requirements are also based on the amount needed to support balance and replace endogenous losses. Zinc is a trace metal nutrient that influences many physiological functions. The first cases of zinc deficiency were described in the 1960s in the Middle East due to consuming plant based diets which contain high levels of antinutrients known to inhibit zinc absorption (Prasad, et al., 1963). The profound effect of zinc deficiency on brain function in both human and experimental animal was reported by Pfeiffer and Braverman (1982) as well as Sandstead et al., 1983). The link between low zinc levels and birth defects, miscarriage, and infection has been reported by several investigators (Chandra, 1980; Hansen et al., 1982;
Liberman and Brunung, 1990). Wallwork et al., (1983) reported the correlation between zinc deprivation and copper accumulation in most regions of the brain while the brain iron was unaffected. Low fetal zinc is also associated with diabetes due to a reduction in placental zinc transfer (Uriu-Hare, et al., 1992). Vegetarians, elderly, athletes and dieters are people with special needs. In the case of pregnancy, women need an additional 3 milligrams daily of zinc. The RDI of iron and zinc are summarised in Table (1).
Table 1: Recommended Dietary Intake (RDI).

<table>
<thead>
<tr>
<th>Groups</th>
<th>Iron (mg/day)</th>
<th>Zinc (mg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infant (0 – 6 months)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Breast- fed</td>
<td>0 – 5</td>
<td>4</td>
</tr>
<tr>
<td>Bottle-fed</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>Infant (7-12 months)</td>
<td>9</td>
<td>4</td>
</tr>
<tr>
<td>Children (1-11 years)</td>
<td>6-8</td>
<td>5</td>
</tr>
<tr>
<td>Adolescents (male or female)</td>
<td>10-13</td>
<td>9</td>
</tr>
<tr>
<td>Adult -male (19-64)</td>
<td>7</td>
<td>12</td>
</tr>
<tr>
<td>(64+)</td>
<td>7</td>
<td>12</td>
</tr>
<tr>
<td>Adult-Female (19-54)</td>
<td>12-16</td>
<td>12</td>
</tr>
<tr>
<td>(54+)</td>
<td>5-7</td>
<td>12</td>
</tr>
<tr>
<td>Post menstruating</td>
<td>5-7</td>
<td>12</td>
</tr>
<tr>
<td>Pregnant</td>
<td>10-20</td>
<td>16</td>
</tr>
<tr>
<td>Pregnant-2&lt;sup&gt;nd&lt;/sup&gt; &amp; 3&lt;sup&gt;rd&lt;/sup&gt; trimester</td>
<td>22-36</td>
<td>16</td>
</tr>
</tbody>
</table>

2.1.22 Mineral inhibitors and enhancers

To absorb a maximum of iron from the food it is necessary to know what enhances as well as what inhibits iron absorption. Several studies have reported that availability of iron is affected by various enhancing and inhibiting factors present in foods (Miller et al., 1981, Hallberg and Rossander, 1982, and Bothwell, 1989).

Components such as certain fibres (Mackler and Herbert, 1985; Brune et al., 1992), polyphenols, phytates, bran, tannic acid or oxalates impair iron absorption by forming more insoluble or undissociated iron compounds (Sandberg, 1991; Brune et al., 1992; Cook, 1997). Factors such as ascorbic acid (Cook et al., 1984; Hunt, 1990 and Fairweather-Tait and Eagles, 1995) and carotene (Garcia-Casal et al., 1998) markedly increase nonhaem iron absorption. Such factors may be considered as increasing the size or availability of the pool of exchange. Siegenberg et al. (1991) stated that ascorbic acid prevents the inhibitory effects of polyphenols and phytates on nonhaem iron absorption. Vitamin C consumed in the same meal may double or triple nonhaem iron absorption (Lieberman and Bruning, 1990; Whitny et al., 1995). This effect is due to a promotion of nonhaem iron absorption (Sayers et al., 1973; Hallberg and Solvell, 1967). Hallberg and Rossander (1984) proposed that the diet in developing countries often has a high content of factors inhibiting nonhaem iron absorption and due to the limited content of food stimulating the absorption of nonhaem iron, the bioavailability of iron is often low. Sharma and
Khetarpaul, (1997) proposed that it is imperative to reduce the level of antinutrients to improve the bioavailability of minerals in the human diet.

2.1.22.1 Phytates

Phytates are a storage form of phosphates. They are salts of inositol hexaphosphate and are a common constituent in grains, seeds, nuts, vegetables and fruits. Phytate accounts for 85% of the total phosphorus stored in many cereals and legumes. In cereals, its distribution varies in that in maize the majority lies in the embryo, while in wheat and rice most of the phytate is found in the aleurone tissue (Kent and Evers, 1993). The level of phytate can vary with type, variety, growing condition, maturity and mill fraction (Lehrfeld, 1989). About 90% of phytate in the western type diet originates from cereals. Phytate binds minerals in the gastrointestinal tract, making dietary minerals unavailable for absorption and endogenously secreted mineral unavailable for reabsorption into the body.

Several studies reported that phytate strongly inhibits iron absorption (Hallberg et al., 1989) and the lower bioavailability of iron in whole meal bread in comparison with white bread has been contributed to the high content of iron phytates in whole meal bread (Hallberg, 1981). Cereals are rich in phytate (Sandberg et al., 1986). Both in vitro and in vivo studies suggested that phytate has
an inhibitory effect on mineral absorption (Navert et al., 1985; Hallberg et al., 1989) because phytate forms insoluble complexes with Fe³⁺, Zn²⁺, Ca²⁺ and Cu²⁺, at physiological pH (Nolan et al., 1987). Phytate has chelating ability. Chelating agents are molecules that coordinate more than once with a metal ion such as iron and zinc (Emsley, 1987). Although the total impact of phytate in the human diet is still not clearly defined, its potential for causing a diet which is marginal with respect to zinc to become truly deficient has been confirmed both in animal and human experiments. Phytate is also one of the determinants of zinc availability especially from cereal based meals and it may act as a zinc depressing agent in human diets (Navert et al., 1985; Feltman 1993).

Cook et al., (1997) reported that phytate content of cereal foods is one of the most important determinants of food iron availability. They also recommend that focus must be on the processing methods used to reduce the content of phytate in cereal. Phytate not only reduces the bioavailability of minerals, but it is proposed that it also affects protein digestibility (Tan et al., 1984). Phytate not only affects the iron absorption from the food in which it is present but also the iron absorption from other foods in the same meal (Layrisse and Martinez-Torres, 1972; Tuntawiroon et al., 1990). Figure 1 and 2 shows phytate molecule as well as chelating ability of phytate respectively.
Figure 1: Phytate molecule.

http://www.apsc.vt.edu/Faculty/Kornegay/sltd001.htm

Phytate Molecule
Figure 2: Chelating ability of phytate.

http://www.apsc.vt.edu/Faculty/Kornegay/skl002.htm

Chelating Ability of Phytate
2.1.22.2 Oxalate

Oxalates are found mainly in foods of plant origin. The actual content varies considerably with season, species, variety, age, maturity and part of the plant (Birch et al., 1972; Williams, 1973). The food sources of oxalate are fruits such as currants, concord grapes, figs, gooseberries, plums, raspberries and strawberries, nuts such as almonds, cashew nuts, and vegetables or herbs such as parsley, beans (green and wax), beets, beets greens, chard, endive, okra, spinach, celery, rhubarb, sweet potato and tomato and beverages such as chocolate, cocoa and tea (Williams, 1973; Thomas and Allen, 1988). The following table (Table 2) presents the amount of oxalates in some foods:

Table 2: Principal sources of oxalate

<table>
<thead>
<tr>
<th>Food</th>
<th>Oxalic acid content (mg/100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beetroot (leaves)</td>
<td>500</td>
</tr>
<tr>
<td>Chocolate/any product containing cocoa</td>
<td>117</td>
</tr>
<tr>
<td>Parsley</td>
<td>100</td>
</tr>
<tr>
<td>Peanuts</td>
<td>187</td>
</tr>
<tr>
<td>Rhubarb</td>
<td>600</td>
</tr>
<tr>
<td>Spinach</td>
<td>600</td>
</tr>
<tr>
<td>Tea</td>
<td>66</td>
</tr>
</tbody>
</table>

Kasidas, 1980
Coenders (1992) proposed that although spinach contains relatively large amounts of iron and calcium, the high level of oxalic acid present in spinach binds these two elements and therefore render them nutritionally almost ineffective. Several studies reported that oxalate or oxalic acid combines with calcium to produce calcium oxalate which is a relatively insoluble compound and therefore prevents calcium absorption (Birch et al., 1972; Williams, 1973). Allen (1982) proposed that although green leafy vegetables such as spinach or other greens contain oxalic acid, these vegetables also contain sufficient calcium to bind its content of oxalic acid and make the oxalic acid unabsorbable, therefore the absorption of iron, calcium or other minerals in other foods eaten at the same time is not likely to be impaired. Wolter et al. (1993) reported a negative influence of oxalic acid on the bioavailability of Zn, Ca and Mg. The very real hazard of oxalate poisoning occurred in Great Britain during World War I when due to the shortage of cabbage the authorities recommended the public to eat boiled rhubarb leaves (Birch et al., 1972). It is well established that the stalks of rhubarb, which are commonly eaten, contain lower concentrations of oxalic acid than the leaves. Robinson et al., (1990) proposed that the occasional use of this kind of vegetables with an adequate intake of calcium is of no concern. Nevertheless it could be a problem if vegetables are eaten frequently and the calcium intake is low.
2.1.23 Mineral fortification

Fortification of iron and zinc especially in cereal products has been practiced in many countries since World War II and is well accepted by food processors and the public. In spite of the fact that iron and zinc fortification is voluntary rather than mandatory (Reilly, 1996) several authorities still use iron fortification to improve the health of their community (Ministry of Agriculture, 1995; National Food Authority, 1994). Reilly (1996) proposed that a large number of manufacturers want to increase the marketability of their foods by adding additional vitamin and minerals to their products.

Iron compounds used to fortify foods may cause organoleptic problems. They result in chemical changes in foods such as: discoloration, acceleration of lipid oxidation, off flavours and metallic taste (Hurrel et al., 1989). Cook and Reusser, (1983) proposed that iron fortification is technically more difficult than fortification with other nutrients because bioavailable iron is chemically reactive and often produces undesirable effects when added into the diet. Ready water soluble iron sources such as ferrous sulfate with highly bioavailable iron can promote rapid fat oxidation and reduce shelf life (Hurrell, 1984; Hurrel et al., 1989). Therefore cereal products with high bioavailable iron are difficult to fortify due to their susceptibility to fat oxidation during storage (Hurrell et al., 1989).
Hallberg and Rossander (1991); Whitney and Rolfes (1993) stated that fortified foods are beneficial when consumed in quantities that meet daily requirements, but at higher levels they can be toxic. The difference between adequacy and excess can be very small. It has been shown that a daily intake of 15 - 17 mg of zinc for which the Australian recommendation is 12 mg per day could alter the body’s copper status, with possible risk of heart disease (Fisher et al., 1984). Excessive zinc intake can impair immune response (Chandra, 1984).

Increasing iron intakes by super enrichment of cereal products may precipitate iron overload in some members of the population (Reilly, 1996). In iron overload, an increasing high level of iron accumulates year by year in the heart, pancreas, spleen, endocrine system, joints and especially in the liver. Researchers in Finland reported that males with the highest level of stored iron have twice the risk for heart attack. Rau, (1998) stated that in iron overload the human body can not reduce the toxic iron excess naturally. Iron overload may be fatal.

The availability of iron in iron fortified foods is reduced by substances that form complexes in the gastrointestinal tract, such as phytate (Fox et al., 1998). Also most compounds used for iron fortification are only partially available for absorption because they are not fully solubilised in the gastrointestinal tract (Hallberg and Rossander, 1991).
Lawrence (1999) stated that taking a soybean gene for the iron storage protein ferritin and transferring this gene into rice is a possible way for producing rice which may contain three times more iron than usual. This proposal raises the question of whether a rice grain with a new gene for the iron storage protein ferritin will taste the same as normal rice or will it taste something between soybean and rice. The higher amounts of iron may change the colour and quality of rice. Also iron rich rice may affect the bioavailability of other minerals. Fisher et al. (1984) proposed that the consequences of excessive intake of minerals have to be considered. It has already been shown that a daily intake of excessive zinc may alter the body's copper status thereby increasing possible risk of heart disease. Also increasing iron intakes by super-enrichment of cereal products would inevitably, precipitate iron overload in some members of the population (Crosby, 1977).

In spite of high amounts of iron and zinc in rice (Hallberg, 1974) bioavailability of these minerals is only about 12% of the total due to the amount of phytate. Phytate content of the rice affects iron absorption from the rice as well as from other foods in the same meal. Approximately 70% of the phytate comes from cereal (Davies, 1981), and by reducing the phytate content of cereal not only the absorption of iron and zinc from cereal may increase but also the absorption of iron from other food in the same meal.
2.1.23.1 Side effects of iron and zinc supplementation:

The RDI which, apply to 95% of individuals are generous recommendations, not minimum requirements, and will usually supply more than the actual needs of individuals. Even with the trace elements that are required in considerably greater quantities, such as iron and zinc, the consequences of excessive intake have to be considered.

Davis and Greger (1992) reported that chronic iron supplementation of 60 mg/day has been associated with low levels of serum manganese after 124 days. Also because iron and zinc are two chemically similar ions, several long term animal and human studies suggest an reverse effect of high Fe/Zn ratios on circulating zinc transport (Prasad et al., 1978; Hamilton et al., 1978). There is an inverse association between circulating levels of zinc and the amount of supplementary iron consumed by pregnant women. Also excess supplemental zinc can cause imbalances of both copper and iron in the body (Patterson et al., 1985).

Hambidge et al., (1983) stated that there is a clear correlation between higher iron intakes by pregnant women in the third trimester with lower plasma zinc level. Although iron supplements can be helpful (Herbert, 1986) for people with a normal iron balance, supplements can promote disability, heart disease, cancer and death (Crosby, 1986 Whitney et al., 1996). Livrea et al. (1996) stated that iron
overload can damage the tissue especially in organs that store iron such as liver. Infections are more likely to occur because bacteria thrive on iron rich blood. Several studies reported that although iron is essential to life in moderate amounts it is toxic in excess quantities (Marshall, 1985; Herbert, 1987; Herbert, 1989).

Unnecessary iron supplements might impair the nutrition of other trace elements and/or impair the immune resistance to infection (Salvioli et al., 1995). Humans are unable to excrete significant quantities of iron and consequently the absorption of iron needs to be controlled (Whitney et al., 1996).

Ferguson et al. (1995) stated that currently supplementation and fortification may face logistical constraints for improving mineral status.

Many research reports have commented upon the adverse effect of excessive or unnecessary iron supplementation, and thus in January 1996, Swedish authorities withdrew totally the legislation requiring mandatory iron fortification of flour.

2.1.24 Dietary factors affecting absorption of iron

Most dietary factors influencing iron absorption exert their action within the
gastrointestinal lumen by making iron more or less bioavailable for absorption (Hallberg, 1998). Dietary constituents that solublise either haem or inorganic iron enhance absorption, whereas compounds that cause precipitation and molecular aggregation decrease absorption. Haem and non haem iron are affected by different compounds due to differences in the availability of coordinating bonds on each moiety (Uzel and Conrad, 1998).

Inorganic iron has six coordinating bonds available for complex formation. Substances such as ascorbic and citric acid bind and alter the physical state of inorganic iron. These components form soluble monomeric complexes with iron, thus preventing polymerisation and promoting absorption. Other chelating compounds, include polyphenols, phosphates, carbonates, tannates and oxalates have an adverse effect on iron bioavailability, forming macromolecules that are insoluble and largely unrecognisable to the intestinal mucosa (British Nutrition Foundation, 1995).

Conversely iron in the haem moiety has only two of its coordinating bonds available for complex formation. Therefore, substances that affect the absorption of inorganic iron have no effect on the absorption of haem iron because they do not alter the physical state of the molecule (Uzel and Conrad, 1998).
2.1.25 Factors which increase iron absorption

Many factors have been identified as influencing the absorption of iron and zinc from vegetables, some enhance such as ascorbic acid and some inhibit such as oxalate. Several studies report that vitamins other than ascorbic acid may enhance the absorption of iron and zinc from a meal.

2.1.25.1 Ascorbic acid

Ascorbic acid or vitamin C was first isolated from lemon juice in 1932. Vitamin C is required for the production and maintenance of collagen, protects against infections and promotes the absorption of iron. In the time of stress, the supply of vitamin C is depleted because it is involved in the release of the stress hormones. This vitamin is also important for the production of thyroxine, the hormone that regulates basal metabolic rate and body temperature (Hamilton and Whitney, 1994). It has been found that the level of ascorbic acid in the diet is an important factor in determining non haem iron absorption (Cook and Monsen, 1977; Monsen et al., 1978). Ramakrishna (1999) stated that iron absorption from dietary sources is based on the ascorbic acid content of the food. Ascorbic acid intake has been found to be more closely correlated to several biochemical parameters of iron nutritional status than was total iron intake (Goralska et al., 1998). Coultate and
Davies (1994) stated that ascorbic acid (vitamin C) enhances the absorption of iron from the small intestine by converting ferric iron to ferrous, whose hydroxide is moderately soluble and can be absorbed. They also mentioned that ascorbic acid in conjunction with the acid contents of fruit forms a loose compound that keeps iron soluble and thus available for absorption.

The Australian RDI for Vitamin C is 60 milligrams for adults, with an extra 20 to 40 milligram recommended for pregnant and lactating women. Citrus fruits are among the best and most popular sources of vitamin C and these can be fresh, canned or frozen. Other vitamin C-rich fruits are strawberries and cantaloupe. The best vegetable sources are broccoli and other members of the cabbage family as well as green leafy vegetables. Tomatoes, potatoes and green pepper have a large amount of vitamin C (Hamilton and Whitney, 1994).

Ball and Barlett (1999) stated that in Australia, both vegetarian and omnivores get approximately 80% of their dietary iron from cereal. Also in many parts of the world the average diet is predominantly cereal in nature with wheat, rice or maize forming the major staples. Although cereals appear to contain sufficient iron (Hallberg, 1982), iron bioavailability is poor in cereals. During digestion the non haem iron in all cereals enter the small intestine where its absorption is influenced by various enhancers or inhibitors. The major known enhancer of iron absorption is ascorbic acid (Derman et al., 1977; Hunt et al., 1990; Walters et al., 1993; Fair weather-Tait et al., 1995). Ascorbic acid owes this ability to its reducing
properties and to its ability to chelate iron. Mac Phail et al. (1981) stated that the promoting influence of ascorbic acid on iron absorption is dependant on its dose.

Kies (1982) stated that to increase non haem iron absorption via ascorbic acid the timing of consumption is very important. Nonhaem iron and ascorbic acid must be consumed at the same time. Considering that important sources of ascorbic acid are all of plant origin, a shift from more animal based foods to more plant based foods should lead to increased consumption of ascorbic acid. However this is not necessarily the case if the shift moves toward a diet based solely, for example, on highly polished cereals.

Gillooly et al. (1983) stated that the ascorbic acid content of the meal should be regarded as the crucial factor in determining its iron nutritive value. An example is the research undertaken by Hallberg (1981) in which the iron absorbed from a vegetarian meal was trebled by addition of cauliflower containing approximately 70 mg ascorbic acid.

2.1.25.2 Vitamin A (β carotene)

The term provitamin A carotenoids is used as a generic descriptor for all
carotenoids exhibiting qualitatively the biologic activity of vitamin A. Both vitamin A and the carotenoids are sensitive to oxidation in the present of oxygen (Wahlqvist, 1998).

Several studies show an association between vitamin A status and iron indices (Mejia et al., 1977; Wolde-Gebriel, 1993). The role of vitamin A on the promotion of inorganic iron absorption was also observed by Layrisse et al. (1997). Although further research is required to confirm this association, it is postulated that vitamin A prevents the inhibition of iron absorption by chemical compounds including phytates and polyphenols. Suharno et al., (1993); Mejia and Chew (1988) stated that to combat nutritional anaemia it is necessary to supplement iron together with vitamin A. Vitamin A can overcome the inhibitory effect of coffee and tea on iron absorption and also may prevent the inhibitory effect of phytates (Layrisse et al., 1997, Garcia-Casal et al., 2000). Garcia-Casal et al. (1998) also stated that vitamin A and β-carotene may form a complex with iron, keeping it soluble in the intestinal lumen, therefore preventing the inhibitory effect of phytates and polyphenols on iron.

2.1.25.3 Riboflavin

Riboflavin (vitamin B2) is a water-soluble vitamin and is found in small amounts in animal and vegetable source foods. Cereals and nut are good vegetable
sources of riboflavin (Wahlqvist, 1998). Riboflavin has the capacity to form complexes and supplementation of riboflavin may result in increased absorption of iron and zinc (Agte et al., 1998). Riboflavin may also limit the accumulation or maintenance of the iron stores (Powers et al., 1983).

Zaman and Verwilghen (1997) proposed that the riboflavin involvement in iron metabolism could be via a flavin dependent oxido-reductase which is capable of removing iron from ferritin.

2.1.25.4 Herbs and iron absorption

Herbs were first written about in technical terms by the ancient Egyptians in approximately 2700 B.C. Herbs were grown and used to promote health (Ortiz, 1993). Herbs are the green or dried leaves of plants and the characteristic aroma and flavour of each herb is due to the essential oil or volatile contents of that herb. Many herbs originated from Asia and South America. The Crusaders and Marco Polo were among the first to introduce herbs to the Western Cuisine (Munro, 1998).
Herbs contain micronutrients. Munro (1998) stated that regular use of herbs or spices can make a significant contribution to the total intakes of minerals such as calcium, iron and zinc. Fresh herbs contain significant amounts of β carotene and vitamin C. Many herbs contain potent antioxidant compounds that provide significant protection against chronic disease (Graig, 1999). Herbs rich in vitamin C or the carotenoids may enhance immune function.

2.1.25.4.1 Dill

Dill (*Anethum graveolens*) with its feathery leaves comes from central Asia and is a very strong aromatic herb. It is believed that the Romans brought this plant to Britain (Stackhouse, 1978). Dill is native to the Mediterranean regions and Southern Russia and is very popular in Iran and Greece as well as Scandinavia where both the leaves and seeds are used but in small quantities (Beckett, 1984; Bremness, 1994). The seeds aid the digestion of vegetables such as cabbage, cucumber and onions and their infusion reportedly reduces stomach pain, hiccups and flatulence (Hemphill and Hemphill, 1990; Toussaint-Samat, 1993; Bremness, 1994).
Herbs contain micronutrients. Munro (1998) stated that regular use of herbs or spices can make a significant contribution to the total intakes of minerals such as calcium, iron and zinc. Fresh herbs contain significant amounts of β carotene and vitamin C. Many herbs contain potent antioxidant compounds that provide significant protection against chronic disease (Graig, 1999). Herbs rich in vitamin C or the carotenoids may enhance immune function.

2.1.25.4.1 Dill

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2.1.25.4.2 Coriander

Coriander (Coriandrum sativum) was grown first in southern Europe. In 1552 B.C. it was believed that coriander could purify the blood and it was
recommended for kidney stones (Hemphill and Hemphill, 1990). It was introduced by the Romans to Britain and then found its way to India, the Middle East, China and South America (Hemphill and Hemphill, 1990). The pungent leaves of coriander are widely used in Middle Eastern and Asian cuisine (Bremness, 1994). Toussaint-Samat (1994) stated that the use of coriander is thought to be typical ingredients of Arab, Chinese and Indian cookery. Coriander has many medicinal virtues, including as a stimulant, a digestive, a carminative and a bactericide.

2.1.25.4.3 Parsley

Parsley (Petroselinum crispum) is probably the best known of the herbs (Toussaint-Samat, 1991). It originally comes from Southern Europe (Ortiz, 1993) and is grown all over the world. The leaves are eaten as a vegetable in the Middle East (Bremness, 1994). It is a plant particularly rich in vitamin A and C, calcium, iron and manganese. A spoonful of chopped parsley a day provides the necessary ration of each of these nutrients (Toussaint-Samat, 1993). The leaves, roots and seeds are diuretic, scavenge skin-ageing free radicals and also reduce the release of histamine (Bremness, 1994).
2.1.25.4.4 Chives

Chives (*Allium schoenoprasum*) belong to the same family as garlic, leek and shallot and are thought to be native of Britain. Bremness (1994) stated that all Allium species contain iron and vitamins and are mildly antibiotic. In the Iranian rice dish (sabzi polow) parsley, coriander and chive or leek are finely chopped and mixed together in equal quantities to flavour and garnish the rice.

2.1.25.5 Vegetables and iron absorption

Vegetables are defined as the fresh parts of plants which, either raw, cooked, canned or processed in some other way, provide suitable human nutrition and can offer almost infinite variety to our dishes. As a food group, vegetables make a valuable contribution to our diet. There are hundreds of different vegetables eaten and available in the Australian market and world wide. Botanically, vegetables are varied as they include the starchy root vegetables such as potatoes and yams, other root vegetables including onions, carrots, parsnips and radishes, the leafy vegetables such as spinach and lettuce, the stalk vegetables such as celery and the flower vegetables such as broccoli and cauliflower (Grosch and Belitz, 1986; Rogers, 1990).
Green leafy vegetables with a very high water content are low in energy and high in vitamin A and C (Coenders, 1992). Vegetables tend to be rich in micronutrients, for example carrots are very rich in β carotene, spinach is rich in iron and folic acid and potato and cauliflowers are very high in vitamin C. Although the absorption of nonhaem iron from vegetables is poor, it is recognised that vitamin C may enhance the absorption of nonhaem iron from vegetables (Truswell, 1998).

Several studies report that the consumption of vegetables may be beneficial for the health. Due to the high antioxidant content, vegetables may reduce the prevalence of certain types of cancer such as bowel, stomach and lung (Truswell, 1998). 2.1.25.5.1 Cabbage

Cabbage (*Brassica oleracea*) is one of the oldest varieties of vegetable. Wild cabbage can still be found growing in many Mediterranean regions and parts of England and Ireland.

Cabbages contain a fair amount of vitamin C with similar quantities of carotenoid, vitamins B and K and calcium and iron (Stackhouse, 1978). Toussaint-Samat (1994) stated that the B vitamins contained in cabbage leaves seem to have soothing and oxygenating qualities.
2.1.25.5.2 Carrot

The carrot (*Daucus carota*) is probably unequalled by any other vegetables as a source of pro-vitamin A and moderate source of vitamin C and fibre. Carrot roots also contain quantities of vitamin B (niacin and pyridoxine). Rogers (1991) proposed that wild carrot originated in Afghanistan. In the twelfth century an Arab writer in Spain wrote about red, juicy and tasty carrots as well as green yellow carrots. In the thirteenth century carrot was known in Italy and in the fourteenth century in France, Germany and Holland. The orange and orange-red varieties are the usual carrot grown, although there are white and yellow carrots. Colour is directly related to the carotene content in carrot, which is fat soluble and is found in the outer layer of the root. Goldberg (1994) reported carrot one of the major food sources of carotene which contain 12 mg/100g carotene.

2.1.25.5.3 Cauliflower

Cauliflower (*Brassica oleracea*) contains large amounts of vitamin C as well as being a good source of folate and a moderate source of iron and fibre. Hallberg and Rossander (1984) stated that the addition of a piece of boiled cauliflower containing 65 mg of ascorbic acid increased the bioavailability of iron in the meal three to four times and while 50 mg ascorbic acid increased it three times. Goldberg
(1994) mentioned raw cauliflower as one of the major food sources of vitamin C while cooked cauliflowers contain 20mg/100gm vitamin C.

### 2.1.26 Nutritive characteristics of vegetables

Vegetables may be an important source of ascorbic acid. They furnish about half of the dietary vitamin A in the form of carotene. Although the bioavailability of minerals such as iron and zinc is low in vegetables, vegetables supply approximately one fifth of the dietary iron in a typical western diet (Robinson et al., 1990). The dark green leafy vegetables are a good source of iron although some vegetables such as spinach and parsley are high in oxalic acid. Several studies have reported that the high oxalic acid in spinach and parsley combines with calcium and/or iron forms insoluble salts that are not absorbed.
Table 3: The nutritional composition of some fresh vegetables, and herbs.

<table>
<thead>
<tr>
<th>mg/100g</th>
<th>Savoy</th>
<th>Cauliflower</th>
<th>Parsley</th>
<th>Coriander</th>
<th>Dill</th>
<th>Lentil</th>
<th>Carrot</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iron</td>
<td>1.1</td>
<td>0.7</td>
<td>7.7</td>
<td>1.9</td>
<td>3.2</td>
<td>11.1</td>
<td>0.2</td>
</tr>
<tr>
<td>Zinc</td>
<td>0.3</td>
<td>0.6</td>
<td>0.7</td>
<td>0.2</td>
<td>1.8</td>
<td>3.9</td>
<td>0.1</td>
</tr>
<tr>
<td>Vitamin C</td>
<td>62</td>
<td>43</td>
<td>190</td>
<td>63</td>
<td>86</td>
<td>Tr</td>
<td>4-6</td>
</tr>
<tr>
<td>Carotene</td>
<td>1.0</td>
<td>0.05</td>
<td>4.04</td>
<td>0.61</td>
<td>6.1</td>
<td>N</td>
<td>43-11</td>
</tr>
<tr>
<td>Cellulose</td>
<td>1.0</td>
<td>0.4</td>
<td>1.5</td>
<td>0.9</td>
<td>1.1</td>
<td>2.9</td>
<td>__</td>
</tr>
<tr>
<td>Soluble fibre</td>
<td>1.7</td>
<td>0.9</td>
<td>2.8</td>
<td>N</td>
<td>1.1</td>
<td>2.0</td>
<td>__</td>
</tr>
<tr>
<td>Insoluble fibre</td>
<td>0.5</td>
<td>0.5</td>
<td>0.7</td>
<td>N</td>
<td>0.3</td>
<td>4.0</td>
<td>__</td>
</tr>
</tbody>
</table>

Holland et al., 1991.

2.1.27 Vegetarian diet and mineral absorption

A well balanced vegetarian diet is compatible with a healthy nutritional status (McLennan and Podger, 1995). Vegetarian dietary practices are associated with a lower incidence of diseases, such as obesity, colon cancer and type two diabetes (Ball and Bartlett, 1999). Some evidence from dietary surveys suggests that there is an inverse correlation between the consumption of leafy green–yellow vegetables and certain disease (Diplock, 1991). There is also evidence that has shown
vegetarian iron intakes to be higher than those of omnivores (Alexander et al., 1994; Kelsay et al., 1988). Nevertheless, both vegetarian and omnivores need to follow dietary practices which enhance iron absorption (Ball and Barlett, 1999).

Vegetables in general contain more iron per calorie than any other foods (Eisman, 1998). Although vegetables contain fibre, oxalate and phytate, which significantly impede absorption of iron in vegetarians, Eisman (1998) stated that a case of iron deficiency anaemia due to plant component inhibition has never been reported. Khader and Rama (1998) stated that the consumption of green leafy vegetables potentially provides the greatest amount of a mineral.

Surveys of vegans and vegetarians have shown that the prevalence of iron deficiency anaemia among vegetarians is similar to the general populations (Latta and Liebman, 1984: Helman and Darnton, 1987).

Mangles (1997) proposed that the satisfactory iron status of many vegans could be due to eating foods which are high in iron as well as a high amount of vitamin C in vegetarian diet. Vitamin C acts to markedly increase absorption of non-heme iron.

Hallberg (1981) stated that adding a vitamin C source to a meal increases non heme iron absorption up to six fold which makes the absorption of non heme
iron as good or even better than that of heme iron. Vegetables such as cauliflower and broccoli which are high in vitamin C are also high in iron thereby enabling the iron in these foods to be very well absorbed. Mangels (1997) stated that eating combinations such as beans and tomato sauce also result in generous levels of bioavailable iron.

2.1.28 Legumes and iron absorption

The legumes are the second most important compartment of the human diet after grain (Mc Gee, 1997). The seeds of legumes are a good source of iron and B vitamins and on average are twice as rich in protein as grains. Dry beans can be substituted for meat to make an economical meal. Berkeley Food Co op (1982) stated that one and one-half cups of cooked beans has the same amount of protein as three ounces of cooked meat. In addition they proposed that beans also provide calcium, iron and B vitamins. With the exception of lentils, most beans need to soak overnight or be boiled in water for a short time, set aside and soaked prior to cooking. Some legumes which are eaten green in the pod, contain more vitamin A and C but less protein.
2.1.28.1 Lentil

The lentil is the oldest cultivated legume (Mc Gee, 1997). Bell (1994) noted that the Greeks and Romans ate large amounts of lentils. It is native in South West Asia and was widely used by Egyptian, Indians and people in Middle East and Central Asia (Toussaint-Samat, 1994). Dried lentils are rich in iron. Toussaint-Samat (1994) proposed that before the seventeenth century the lentil soup provided for the Roman legions by the consuls sustained their iron nutriture.

2.1.28.2 Mung bean

Green and golden mung bean from the Fabaceae family are cultivated in Asia for their nutritious dried seeds (Smith, 1998). Most of the world’s mung bean are grown and eaten in India as well as in China and Asia where it is used for flour and boiled, mashed dishes as well as sprouts (Mc Gee, 1997). Mung beans are consumed primarily as sprouts in Western cuisine.
2.1.29 The effect of processing on vegetables

Different processing techniques gives vegetables a longer storage capacity compared with fresh vegetables. Processing including canning often has an negative effect on vitamins. Belits and Grosch (1987) proposed that canning of the vegetables moderately destroyed (5-30%) carotene. Vitamin B1 in carrots and tomatoes does not decrease significantly where as in some other vegetables such as green beans, peas and asparagus the losses are 10-50%. Due to enzymatic and chemical digression, vitamin C retention is 55-90% during canning of asparagus, peas and green beans.

2.1.29.1 Effect of freezing on vitamin and mineral of vegetables

Freezing reduces all biochemical and respiratory activities to a minimum. During freezing, cellular water osmotically pass as through the cell wall into the intercellular spaces where it freezes to form solid crystals. The faster this happens, the smaller the crystals and consequently the smaller the mechanical damage to the cell wall. Losses of vitamins or minerals may occur during the preparation for freezing such as washing, blanching, grinding or trimming. Vitamin C losses are
specially likely, because they occur whenever tissues are broken and exposed to air. Uncut vegetables or fruits do not lose their vitamin C. Hamilton and Whitney (1994) stated that freezing itself does not destroy nutrients. The nutritional losses are small in comparison with those that occur during thawing and cooking (Wahlqvist, 1998). Olson and Dietrich (1969) stated that, with the good freezing technique, there is not much difference between frozen vegetables and those only cooked. To have a longer storage life without losing the nutrients, Hamilton and Whitney (1994) stated that foods need to be kept at a temperature colder than 18°C or 0°C. Carrot is a suitable vegetable for freezing. Margen (1992) proposed that the nutritional differences between frozen and fresh carrots are generally minor although the fresh carrots have approximately twice as much vitamin C as the frozen one.

2.1.29.2 Effects of dehydration on nutrient content of vegetables

Dehydration is the removal of water content by the application of heat. Therefore, in dry food the residential water concentration is too low to support the growth of micro-organisms. Sun-drying is an ancient method which is applied to some fruits and vegetables in the hotter climates. It is still used in some parts of Asia with abundant sunlight to dry vegetables, fruits, meat and fish (Wahlquist, 1998). Depending on the range of temperature uses and conditions, the nutrient losses are very varied. Wahlquist (1998) stated that sun-drying and air drying cause more nutritional damage than spray drying or freeze drying with vitamin C and the vitamin B group being most affected.
CHAPTER 3

SURVEY OF THE RICE CONSUMPTION, PRECOOK TREATMENTS AND THE METHOD OF COOKING BY IMMIGRANTS IN SYDNEY.

3 INTRODUCTION

Pattern of food selection, distribution and consumption vary in different types of societies. The method of preparation may carry nutritional implications in terms of nutrient retention or loss. For example, "The specific mix or combination of staple grains, seeds and legumes is important to the protein content of the diet" (Jerome et al., 1980). Also Berlyne et al, (1973) proposed that osteomalacia in the Bedouinian females living in the Negev Desert of Israel is due to the consumption of unleavened bread which contains high amounts of phytic acid.

Mineral absorption may be inhibited or enhanced with certain food combinations. For example, iron absorption is enhanced in the presence of ascorbic acid or inhibited in the presence of phytate. The addition of certain ingredients during food preparation may modify the nutritional composition of the food. "Traditional alkali processing of maize in the preparation of tortilla increases the availability of lysine in its overall nutritional quality" (Jerome et al., 1980).
3.1 The objectives of this survey

- To determine Iranian immigrants' consumption patterns of rice in terms of how often rice is consumed.

- To understand the main factors affecting rice consumption, particularly any change in the consumption patterns as a result of immigration.

- To determine the traditional rice preparation methods in terms of precooking treatments.

- To determine the traditional method of cooking rice.

- To identify the traditional rice dishes consumed including the vegetables, herbs and legumes used in the rice dishes.
3.2 Material and Methods

3.2.1 Method

Numerous methods for measuring dietary intakes of individuals and groups exist (Fong and Kretsch, 1990). The method used for collecting information about rice consumption patterns for this exploratory research was by means of survey questionnaire. Iranian migrants to Australia were chosen as the subjects due to the fact that these people come from an area where iron and zinc deficiency is common. In addition, rice is one of their dietary staples.

3.2.2 Designing of questionnaire

The questionnaire was designed according to Patton (1982). Patton suggested that for designing a questionnaire the following points need to be addressed.

- What do you want to find out? This question seems simple but often requires hours of discussion to arrive at an answer.

- Why do you want to know? This is the purpose of the study.
- When do you need the information? A deadline will help in developing a timeline for any studies. Consider how long it will take to design the study, develop the questioning route, take care of logistics, recruit participants, analyse the data and prepare the report.

- How can you get the information needed? Interviewing could be an appropriate method to obtain the kind of information needed. With interviewing it is possible to determine how a percent of population consumes or prefers food as well as their cooking procedures.

- Where do you get the information? The geographic locations and the specific sites need to be considered. Where are the people who can provide the information we need? Are they in certain areas? Due to the fact that the aim of most investigations is to generalise the results from a specific group to the population as a whole, it is important to have normative data on both the population and the study group.

- From whom should you collect the information? Thinking about who can provide the needed information is an important part of any survey. Therefore demographic characteristics of participants must be considered, for example gender, age, income, ethnic background, type of household and location of residence.

Prior to the survey, the dietary questionnaire was pretested on fifteen subjects to assess acceptability of the length and format. After the questionnaire was revised and approved by the UWSH Human Ethics Committee (Aug.1998) the survey was conducted. The interview was undertaken after obtaining a consent form. The
questionnaire took twenty to thirty minutes to complete. Most of the questions were close ended. On the average six to eight subjects were interviewed per day. Data were collected from July to September 1998.

3.2.3 Method of data collection

The introductory page of the questionnaire explained the reason for survey as well as containing the informed consent form in compliance with the University of Western Sydney, Hawkesbury, Human Research Ethics Committee Guidelines. The questionnaire contained twenty-one questions consisting of both open and close-ended questions and was administered in an interview format which began with demographic data. Several questions were directed at determining the method of precooking and cooking of rice by this population. Additional questions looked at eating habits as well as specific rice dishes.

The first part of the questionnaire was comprised of simple biographical questions which assessed age, level of education and employment status. These questions were asked to elicit basic background information.

The second part was a series of open and close-ended questions to determine the rice consumption patterns, the type of traditional precooking treatments and the overall cooking method of rice.
The third part assessed the perceived change in rice consumption patterns as a result of immigration.

The fourth part of the questionnaire was designed to determine traditional rice dishes consumed as well as the kinds of herbs, vegetables and legumes added to the rice dishes.

The last few questions give information about food consumption frequency as well as the nutrition knowledge of the population concerning mineral deficiencies. Food frequency questionnaires were used to assess the food acceptance and rejection. It included fresh, dried and canned fruit and vegetables as well as meat, rice and drinks. These foods are expected to be used with varying frequency, both high and low. All foods are readily available in the Sydney area. See the copy of the survey questionnaire in appendix 1.

3.2.4 Sources of error in survey

Provision of reliable data depends upon the willingness, motivation, and knowledge of the study purpose and memory of the participants. This problem may be minimised by thoroughly explaining the purpose of the study.
Interviewer’s reactions or opinions may also influence the subject’s responses. Witschi (1990) stated that a non-judgemental attitude towards the respondent is essential. A neutral reaction towards the respondent responses is necessary.

Finally to prevent any systematic errors caused by incorrect coding all data were checked twice.

3.2.5 Definition of subjects and sampling frame

Subjects consisted of 107 adult female-immigrants from Iran, who were presently residing in Sydney. Subjects were contacted by phone and were selected at different Iranian cultural meetings such as New Year celebration, Iranian Welfare Association and the Islamic prophet’s birthday celebration. All participants were bilingual, speaking at least English and Farsi. They were informed that they had the right and option to withdraw at any time during the interview.

3.2.6 Significance of the study

This research will provide information about the traditional method of preparation and cooking as well as consumption pattern of rice. The awareness of Iranian immigrants about iron and zinc deficiency and the mineral contents of foods are also assessed.
The rice consumption and preparation patterns are examined to determine the most frequent method of preparation and cooking of rice. It may also be used for the development of new rice preparation methods for retention of maximum amounts of different nutrients in rice. The following mind map briefly illustrates the intention of this study.

3.2.7 MINDMAP
3.3 Analysis of data

After collecting the information, data was coded and analysed using the statistical package (SPSS) and computer program, Microsoft Excel 97. The computer used was an IBM compatible PC.

For analysis of data the following variables were coded and analysed:

- Age and cooking method
- Education and cooking method
- Employment history
- Frequency of rice consumption
- Rice precook treatments
- Rice cooking methods
- Rice dishes consumed
- Frequency of rice dishes consumed
- Perceived change in rice consumption patterns
- Reason for consumption of rice
- Concern about the brand of rice
- Location of purchasing rice
- Frequency consumption of food items
- Awareness of iron and zinc nutriture
- Awareness concerning iron and zinc deficiencies
3.4 Results and discussion

3.4.1 Demographic data

The intercept survey produced the following demographic data (Table 4, 5 and 6). Of the 107 respondents 6% were under 25 years, 33% were 26 to 35 years and 43% were 36 to 45 years with 18% being more than 46 years old. Only women were selected with most (82%) being educated at the secondary level or higher. Fifty three percent were employed. Rahmanifar and Bond (1990) stated that in middle and high-income families in Iran most women have high school certificate.

Table 4: Distribution of the subjects by age.

<table>
<thead>
<tr>
<th>Age</th>
<th>Numbers</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Under 25</td>
<td>6</td>
<td>5.6</td>
</tr>
<tr>
<td>26-35</td>
<td>35</td>
<td>32.7</td>
</tr>
<tr>
<td>36-45</td>
<td>46</td>
<td>43</td>
</tr>
<tr>
<td>46+</td>
<td>20</td>
<td>18.7</td>
</tr>
<tr>
<td>Total</td>
<td>107</td>
<td>100</td>
</tr>
</tbody>
</table>
Table 5: Percent distribution of subjects according to their level of educations.

<table>
<thead>
<tr>
<th>Educational level</th>
<th>Number</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Primary school</td>
<td>16</td>
<td>15</td>
</tr>
<tr>
<td>Secondary school</td>
<td>33</td>
<td>31</td>
</tr>
<tr>
<td>University/College</td>
<td>55</td>
<td>51</td>
</tr>
<tr>
<td>Total</td>
<td>107</td>
<td>100</td>
</tr>
</tbody>
</table>

Table 6: Percent distribution of subjects according to their employment status.

<table>
<thead>
<tr>
<th>Employment status</th>
<th>Numbers</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Employed full time</td>
<td>27</td>
<td>25</td>
</tr>
<tr>
<td>Employed part time</td>
<td>30</td>
<td>28</td>
</tr>
<tr>
<td>Unemployed</td>
<td>43</td>
<td>40</td>
</tr>
<tr>
<td>Student</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>Total</td>
<td>107</td>
<td>100</td>
</tr>
</tbody>
</table>

Several factors may determine food habits and behaviour of individuals or groups. Wahlquist (1998) and Stanton (1990) believe that culture, religion, time and cooking skills as well as cost are all factors which influence individual food behaviour.
Factors such as age, educational level and employment status might affect the food behaviour of individuals. Examining the cooking method and age (Table 7) in this survey shows that the subjects overall (67%) preferred to use the boiling method for rice preparation, although the absorption method gained favour with the older subjects.

Employment status as well as the educational level did not influence the rice cooking behaviour of the subjects.

Table 7: Correlation between age and the method of cooking rice used by subjects.

<table>
<thead>
<tr>
<th>Age</th>
<th>Absorption method (%)</th>
<th>Boiling method (%)</th>
<th>Others (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Under 25</td>
<td>3</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>26-35</td>
<td>5</td>
<td>26</td>
<td>2</td>
</tr>
<tr>
<td>36-45</td>
<td>10</td>
<td>32</td>
<td>1</td>
</tr>
<tr>
<td>46+</td>
<td>12</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>30</td>
<td>67</td>
<td>3</td>
</tr>
</tbody>
</table>

N = 107

Figure 3 illustrates the popular methods of cooking rice in the Iranian culture.

Overall boiling is the most popular method of cooking rice.
Figure 3: Distribution of subjects who cook their rice with different methods (%).
3.4.2 Cooking

Women with families may spend a large amount of time on preparation and cooking of food. A range of factors influence food selections. Convenience seems to be a major determinant (Thomson, 1988). Although it is simplistic to say that good or poor nutrition depends on nutrient intake, it must be remembered that good or poor nutrition is often the result of the food choices people make as well as the cooking method. For example, the method of cooking may enhance or decrease the real and/or the perceived nutritional image of a food. Frying of fish damages its image as a healthy food while poaching enhances it (Thomson, 1988).

The quality of rice as a food is dependent on the way that it is cooked (Marsh and Wadsworth, 1994). Different methods of cooking produce different textures and appearances. Nutritionists need to be aware that individual food behaviour reflects a person's relationship to food throughout life (Bass et al., 1979).

Rice is a popular and widely consumed food in the Iranian culture (Rahmanifar and Bonds, 1990). Results from this survey show that the large majority of participants cook rice either every day or at least 3 to 4 times a week (Figure 4).
Figure 4: Frequency of rice cooking.

N= 107
The majority of the subjects reported that they consumed rice because they like it (32%). Rozin and Voilmecke (1986) proposed that food likes and dislikes are a very powerful force in human nutrition.

Fieldhouse (1995) stated that food may be accepted even though it is not preferred, for reasons of availability, cost or social courtesy. Tradition may regulate the pattern of exposure to foods, the nature of foods, and their flavouring and preparation whereas we may choose different foods when our favourite food is hard to reach, expensive or difficult to prepare. In this survey, nutritional value and tradition were factors affecting the subjects preference for consumption of rice whereas cost had less effect on their selection (Figure 5). Axelson (1986) also stated that the impact of culture on food preference is extremely large.

Since rice is a major part of Persian cuisine naturally, Iranians are often adamantly particular about the quality of rice they choose. The majority of the subjects reported to purchasing rice from either Iranian (41.7%) or Middle Eastern (43.7%) grocery stores. Ghanoonparvar (1982) stated that Middle Eastern grocery stores might offer the closest rice substitute in flavour.
Figure 5: Perceived reason for consumption of rice by Iranian.

Reasons

- Like it
- Nutritional value
- Tradition
- Availability
- Convenience
- Cost
The main purpose of this survey was to obtain information about the method of preparation and cooking of rice. From this survey, it was found that the majority of subjects wash and soak rice prior to cooking (Figure 6). Although washing rice may not be really necessary, it is often recommended that rice not be washed in order to retain any enrichment. However in the Iranian culture, washing is an unavoidable part of the proper preparation of rice. Figure 7 shows the frequency of washing rice by the Iranian subjects.
Figure 6: Distribution of subjects who wash or soak their rice before cooking.

N: 107
Figure 7: Methods of washing rice (%).

No soaking: 50%
Plain water: 29%
Salted water: 21%

N: 107
The majority of subjects reported using salted water to soak rice prior to cooking (Figure 8). Khalili Batmanglij (1992) stated that soaking and cooking in salt water helps firm up the rice, lengthen it, and keep it separated and fluffy after the cooking process. Basmati rice is washed thoroughly in cold water by placing rice in a large pot and covering with cold water, agitated briskly by hand, and pour off the water. This procedure is repeated five times until the rice is completely clean (Khalili Batmanglij, 1992). Khalili Batmanglij (1992) also proposed that in Northern Iran (Gillian) rice would often be soaked in salt water in large quantities and used for cooking as needed. The soaking time varies between less than 2 hours to a maximum of 24 hours. Khalili Batmanglij (1992) said that it is desirable to soak rice in salted water for the period of 2 to 24 hours. She recommended that cooking of rice in salted water support the long cooking time as well as preventing the breaking of the rice grain. The majority of subjects soaked their rice for up to 2 hours (Figure 9).

Figure 8: Frequency of rice soaking procedure.
Figure 9: Distribution of rice soaking time prior to cooking.

N: 107
3.4.3 Rice dishes consumed by Iranians

To obtain information about the rice dishes usually consumed by Iranians, the subjects were asked how they serve their rice dishes as well as to specify the herbs, vegetables and legumes they may use in rice dishes. The majority of the subjects reported consuming plain rice with either stew, red meat or white meat 3 to 4 times per week. Table 8 shows the frequency of rice dishes consumed by Iranians. The most popular rice dish included dry dill and broad beans and usually was served with lamb meat. A mixed rice dish with cauliflower and minced lamb was also popular. The frequency consumption of plain rice (rice consumed without any other foods) was only 4% on a weekly basis. Therefore as the majority of the subjects reported to cook rice on a daily basis (Figure 4), they usually consume rice either with stew, white meat, red meat or as a rice dish with vegetables, herbs or legumes. Ghanoonparvar, (1982) stated that rice is certainly one of the most important staples in the Iranian diet. He also mentioned rice as a main dish and that other foods or ingredients are put in it, over it or around it. The herbs, vegetables and legumes used by this community in rice dishes are listed in Table 9.
Table 8: Frequency of serving rice dishes (%).

<table>
<thead>
<tr>
<th></th>
<th>Daily</th>
<th>4 to 5/week</th>
<th>Weekly</th>
<th>Rarely</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plain rice</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>0</td>
<td>96</td>
</tr>
<tr>
<td>Plain rice with stew</td>
<td>0</td>
<td>48</td>
<td>36</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td>Plain rice with white meat</td>
<td>0</td>
<td>35</td>
<td>52</td>
<td>12</td>
<td>1</td>
</tr>
<tr>
<td>Plain rice with red meat</td>
<td>0</td>
<td>56</td>
<td>39</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td>Mixed rice with vegetables/herbs</td>
<td>0</td>
<td>16</td>
<td>38</td>
<td>42</td>
<td>4</td>
</tr>
<tr>
<td>Mixed rice with legumes</td>
<td>0</td>
<td>18</td>
<td>34</td>
<td>39</td>
<td>9</td>
</tr>
</tbody>
</table>

Table 9: Herbs, vegetables and legumes used in Iranian rice dishes.

<table>
<thead>
<tr>
<th>Herbs</th>
<th>Vegetables</th>
<th>Legumes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coriander (fresh, frozen or dry)</td>
<td>Savoys/Cabbage (Fresh or frozen)</td>
<td>Broad beans (Dry)</td>
</tr>
<tr>
<td>Parsley (Fresh, frozen or dry)</td>
<td>Carrot (Fresh)</td>
<td>Lentils (Dry)</td>
</tr>
<tr>
<td>Dill (Fresh frozen or dry)</td>
<td>Cauliflowers (Fresh or frozen)</td>
<td>Green beans (Fresh or frozen)</td>
</tr>
<tr>
<td>Chive (Fresh or frozen)</td>
<td></td>
<td>Eye bird beans (Dry)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Mung beans (Dry)</td>
</tr>
</tbody>
</table>

Table 8 shows the distribution of subjects who add herbs, vegetables and legumes to their rice dishes. Among herbs, dill was the most popular in these Iranian

95
rice dishes. The most popular vegetable was cauliflower, which was reported to be added weekly to the rice dishes by the majority of the subjects.

3.4.4 Food consumption and dietary pattern

Food frequency of major food items was determined. Food items such as brown rice, canned fruit and vegetables and fish were used rarely while fresh fruit and vegetables were consumed daily by most of the subjects, 88% and 64% respectively (Table 10). All subjects generally followed a traditional Iranian food pattern although differences in food preferences and consumption were evident.
Table 10: Food consumption pattern (%).

<table>
<thead>
<tr>
<th></th>
<th>Daily</th>
<th>2-3/Week</th>
<th>Weekly</th>
<th>Rarely</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Fruit</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fresh</td>
<td>82.7</td>
<td>7.2</td>
<td>2.5</td>
<td>2.5</td>
<td>0</td>
</tr>
<tr>
<td>Dried</td>
<td>26.8</td>
<td>0</td>
<td>19.5</td>
<td>43.9</td>
<td>9.7</td>
</tr>
<tr>
<td>Canned</td>
<td>0</td>
<td>4.9</td>
<td>9.7</td>
<td>61</td>
<td>24.4</td>
</tr>
<tr>
<td><strong>Vegetables</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fresh</td>
<td>65.9</td>
<td>14.6</td>
<td>12.2</td>
<td>7.3</td>
<td>0</td>
</tr>
<tr>
<td>Dried</td>
<td>2.5</td>
<td>9.7</td>
<td>17.1</td>
<td>41.5</td>
<td>29.2</td>
</tr>
<tr>
<td><strong>Meats</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Red</td>
<td>46.4</td>
<td>34.1</td>
<td>12.2</td>
<td>7.3</td>
<td>0</td>
</tr>
<tr>
<td>White</td>
<td>14.6</td>
<td>53.7</td>
<td>26.8</td>
<td>4.9</td>
<td>0</td>
</tr>
<tr>
<td>Fish</td>
<td>2.5</td>
<td>24.3</td>
<td>34.1</td>
<td>36.6</td>
<td>2.5</td>
</tr>
<tr>
<td><strong>Rice</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>61</td>
<td>26.8</td>
<td>12.2</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Brown</td>
<td>0</td>
<td>0</td>
<td>4.9</td>
<td>17.1</td>
<td>78</td>
</tr>
<tr>
<td><strong>Drinks</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tea</td>
<td>90.2</td>
<td>7.3</td>
<td>2.5</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Coffee</td>
<td>46.3</td>
<td>14.6</td>
<td>2.5</td>
<td>24.4</td>
<td>12.2</td>
</tr>
<tr>
<td>Water</td>
<td>65.8</td>
<td>4.9</td>
<td>2.5</td>
<td>19.5</td>
<td>7.3</td>
</tr>
</tbody>
</table>

Dark tea was the most popular drink consumed by the subjects and rice and bread were the staple foods. If one meal included rice, the other did not. These results are in agreement with previous reports on Iranian food patterns (Rahmanifar and Bond, 1990),
Some of the subjects believed that their rice consumption had decreased since immigrating to Australia (Figure 10), but nearly half reported that it remained unchanged.

Rice was purchased from a variety of shops. Iranian shops accounted for the largest single site of rice purchasing with ethnic grocery stores also being widely used.

Figure 10: Perceived change in the consumption of rice since immigration to Australia.
3.4.5 Nutrient knowledge of the subjects

In response to the question asking what is a good source of iron or zinc the majority of subjects believed that vegetables and red meat (75.7%, 73.8% respectively) are a good source of iron. Very few responses indicated a good understanding of the amount of absorbable iron or zinc in food (Table 11 and 12).

Table 11: Subjects responses and consideration of a good source of iron.

<table>
<thead>
<tr>
<th>Foods</th>
<th>Yes</th>
<th>No</th>
<th>Do not know</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>Red meat</td>
<td>79</td>
<td>73.8</td>
<td>3</td>
</tr>
<tr>
<td>White meat</td>
<td>18</td>
<td>16.8</td>
<td>33</td>
</tr>
<tr>
<td>Vegetables</td>
<td>81</td>
<td>75.7</td>
<td>3</td>
</tr>
<tr>
<td>Fruits</td>
<td>53</td>
<td>49.5</td>
<td>18</td>
</tr>
<tr>
<td>Cereals</td>
<td>48</td>
<td>44.8</td>
<td>15</td>
</tr>
<tr>
<td>Pulses</td>
<td>48</td>
<td>44.8</td>
<td>15</td>
</tr>
</tbody>
</table>

No. = 107

The least number of correct responses were received for the question regarding dietary zinc. The survey answers indicated that the information with regards to dietary source of zinc is very limited. A summary of the responses is presented in Table 12.
Table 12: Subjects responses and consideration of a good source of zinc.

<table>
<thead>
<tr>
<th>Foods</th>
<th>Yes</th>
<th></th>
<th>No</th>
<th></th>
<th>Do not know</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>Red meat</td>
<td>18</td>
<td>16.8</td>
<td>10</td>
<td>9.3</td>
<td>79</td>
</tr>
<tr>
<td>White meat</td>
<td>16</td>
<td>14.9</td>
<td>10</td>
<td>9.3</td>
<td>81</td>
</tr>
<tr>
<td>Vegetables</td>
<td>17</td>
<td>15.9</td>
<td>5</td>
<td>4.7</td>
<td>85</td>
</tr>
<tr>
<td>Fruits</td>
<td>19</td>
<td>17.7</td>
<td>5</td>
<td>4.7</td>
<td>83</td>
</tr>
<tr>
<td>Cereals</td>
<td>18</td>
<td>16.8</td>
<td>5</td>
<td>4.7</td>
<td>84</td>
</tr>
<tr>
<td>Pulses</td>
<td>19</td>
<td>17.7</td>
<td>5</td>
<td>4.7</td>
<td>83</td>
</tr>
</tbody>
</table>

In response to the question of using vitamin or mineral supplements, 77.5% of the subjects reported no use of such supplements (Figure 119).

Although the majority of the subjects reported feeling tired as well as reducing exercising, about 7% reported having either mild or severe anaemia (Figure 12).
Figure 11: Distribution of the subjects who use supplements.

Supplements
- Calcium
- Iron
- Vitamin
- Folic acid
- No pills

78% 7% 5% 3% 7%
Figure 12: Subjects perceived of feeling or having the followings.

- 30% Energetic
- 27% Tired
- 14% Lethargic
- 10% Reduced ability to work
- 4% Poor stamina
- 15% Anaemia
3.4.6 Cookware

Using different materials as a cookware may present different health issues (Upton et al, 1993). Over 60% of the subjects reported using non-sticking pans for cooking rice. Upton et al., (1993) proposed that such pans require less fat for cooking and can be cleaned easily and quickly. Stainless steel and aluminium pans were used much less frequently. Concern about aluminium leaching into the food is unwarranted according to Upton et al., 1993. Table 13 shows the kind of pan usually used for cooking rice and rice dishes.

<table>
<thead>
<tr>
<th>Cookware</th>
<th>No.</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stainless steal</td>
<td>20</td>
<td>18.7</td>
</tr>
<tr>
<td>Non stick</td>
<td>67</td>
<td>62.6</td>
</tr>
<tr>
<td>Aluminium</td>
<td>15</td>
<td>14</td>
</tr>
<tr>
<td>Others</td>
<td>5</td>
<td>4.7</td>
</tr>
<tr>
<td>Total</td>
<td>107</td>
<td>100</td>
</tr>
</tbody>
</table>

Table 13: Frequency of subjects using different kinds of pan for cooking rice.
3.5 CONCLUSION

The purpose of this chapter was to determine the food habits and the cooking methods of rice by Iranian immigrants living in Australia (Sydney). The information gained from this chapter will be used to ascertain the amount of total and bioavailable iron and zinc as well as phytate content in the Iranian rice dishes.

Rice is one of the staples in Iran and 83% of the subjects reported either cooking rice every day or 3-4 times per week. The majority of the subjects reported that they wash rice before cooking and 50% soak their rice in salted water. The soaking time was reported to be in the range between less than 2 hours to 24 hours with the majority soaking rice for less than 2 hours.

Analyses of the data showed that the most preferred cooking method was boiling with 67% reported cooking rice via boiling method and 30% reported to using the absorption method. Only 3% used some other method for cooking rice.

Besides cooking and using plain white rice the majority of the subjects reported cooking a variety of rice dishes including rice with either herbs, vegetables or legumes. The most popular rice dishes were rice with dill and broad bean as well as with cauliflower. Both were reported to be consumed weekly by the majority of the people.
CHAPTER 4

MEASUREMENTS OF TOTAL AND BIOAVAILABLE IRON AND ZINC AS WELL AS PHYTATE IN BASMATI RICE.

4 INTRODUCTION

Rice is the staple food for a majority of the world’s population (Tuntawiroon et al., 1990). Although rice is sufficient in minerals such as iron and zinc, its low bioavailability is one of the most significant factors for deficiency of these elements (Indumadhavi and Agte, 1992). Several studies suggest that iron and zinc absorption are low from rice based meals. One of the major inhibiting factors in cereal based food for zinc and iron absorption is phytate phosphorus (Hung and Lantzsch 1983; Sandberg 1991; Cook et al. 1997). Phytate may inhibit several proteolytic enzymes (Knuckies et al. 1985) and interact with protein metabolism. During proteolytic digestion, phytate may decrease the availability of proteins (O’Dell and Boland 1976). Although rice contains essential iron and zinc, the bioavailability of these elements is only about 10% of the total (Mills et al. 1988). DeMaeyer and Adiels-Tegman (1985) reported that the prevalence of iron deficiency is especially high in countries where there is high dietary rice intake. Iron and zinc are essential micronutrients and a cheap and acceptable method is required to improve their bioavailability in the diet.
Traditional household methods such as the particular soaking and cooking method can have both positive and negative effects on mineral availability.

4.1 MATERIALS AND METHODS

This chapter describes the chemicals, equipment and experimental methods that were used to measure the total and bioavailable iron and zinc as well as phytate in two cultivars of rice. Basmati rice was the rice of choice in Iran and the brown rice has used for comparison sake due to its high phytate and fibre contents.

4.1.1 Samples

Two cultivars of rice, Basmati (white rice, 5kg) and Sunbrown Natural Calrose (brown rice, 5kg) were procured from a local market in Sydney and stored in airtight dishes at room temperature.

In order to check the degree of polishing of both cultivars the rice producers (Rice Growers for Sunbrown rice and Riviana Foods for Basmati rice) were contacted. In response, Ricegrowers' Co operative Limited-Leeton (personal communication, 1997) stated that Sunbrown has had the rice hulls removed only but has not been polished. Riviana Foods responded that the company only imports polished rice and had no information about the degree of polishing of the Basmati
rice. The Basmati rice was imported from Pakistan and is similar to the rice consumed in Iran and other Middle Eastern countries.

4.1.2 Spectrophotometer

Absorption spectrophotometry is common spectroscopic method. The SpectrAA OS/2 Varian spectrophotometer used in this experiment for measurements of phytate and phosphorus contents of samples is controlled via RS232C interface from an IBM computer having a 3.5 inch disc drive with user friendly PC software operating in the Microsoft Windows environment (version 3.0). Due to the fact that this is the method of choice for this study, it will to be reviewed in more detail.

The spectrophotometric method is based on Beer's law, which proposed that the absorption of a medium directly depend on the medium and its surroundings (Strobel and Heineman, 1989). Light of power P passes through an infinitesimally thin cross-section of a sample containing dn absorbers and loses a fraction of its power dp / p. These quantities can be connected by the equation: \( -\frac{dp}{p} = k \, dn \) where k is a proportionality constant. This differential equation can be integrated between the limit of \( p_0 \) to \( p \) and of \( k \, dn \) between 0 and \( n \), yielding the following equation: \( \ln \frac{p}{p_0} = -kn \) with \( p_0 \) as the power incident on the sample, \( p \) as the power leaving the sample and \( n \) related to the concentration. The number of absorbers in the beam must be expressed in terms of concentration. The following equation gives you the number of absorbers in the beam: \( N = c_0 \, N_{\text{abs}} \) with c as the
concentration of absorbers, $b$ as the thickness of the cell, $N_0$ as Avogadro's number and $s$ as the cross sectional area of the beam. For $C$ in units of mol L$^{-1}$ and $b$ in cm, conversion of $\ln p/p_0 = -kn$ to the Naperian logarithm to base 10 designated by $\log$, and the concentration is changed to units of molarity: $\log p_0 / p = A = \epsilon bc$ with $A$ as absorbance and $\epsilon$ as the molar extinction coefficient (L mol$^{-1}$ cm$^{-1}$). To determine the number of molecules in a sample in a given cell absorbing at a particular wavelength, a change in the absorbance of the sample at the particular wavelength following irradiation can be used in conjunction with the equation:

$$\log p_0 / p = A = \epsilon bc.$$  

4.1.2.1 Optical description

The diagram shows in a simplified form the optical system of the instrument. Light from either the Deuterium or Tungsten lamp passes through the sample, which absorbs some of the UV or visible light. The remaining light passes onto the diffraction grating in the monochromator before being passed onto the photomultiplier tube detector to produce a signal which is then amplified and displayed (Figure 13).
4.1.2.2 Cleaning of spectrophotometric cells

Spectrophotometric cuvettes with a path length of 10 mm and a volume of 4.0 ml were used in this experiment. Such cuvette surfaces tend to become contaminated easily, and gross contamination may occur from dust, finger marks, grease and sample evaporation. Such contamination decreases the transmission and may contaminate the samples. Thus, care was taken to ensure that the cells were cleaned thoroughly before each experiment. The procedure used to clean the cells was adapted from the
method in AOAC, (1990). The cells were immersed and left for 30 minutes in freshly prepared nitric acid (1:1) and then rinsed four times with distilled water and air dried.

4.1.3 Atomic absorption spectrophotometer

Atomic absorption spectrometer has been used for analysis of trace elements for many years and it is at the forefront of trace metal technique (Ewing, 1990). Allan and David were the first to use atomic absorption spectrophotometer (AAS) to determine the amount of iron and zinc (David, 1958; Allan, 1958). The atomic absorption procedure is an accurate and reliable method for measuring trace elements, especially iron because iron determination is almost free of any interference when an air acetylene flame is used (Salvin, 1968). Cresser, (1987) proposed that AAS has a detection limit for iron of 0.01 μg ml⁻¹. The AAS determination of zinc is also highly selective and sensitive with a detection limit of 0.002 μg ml⁻¹. The atomic absorption process is illustrated in figure 14:

Figure 14: The process of atomic absorption spectrophotometer.

Lamp → Flame → Monochromator → Detector

\[ V \xrightarrow{10} \text{ШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШШŠ

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The absorption of a photon of specific energy that corresponds to the difference between the lower and upper energy levels of a free atom in the hot sample gas yields the AAS measurements. \( I_0 \) is the light at the resonance wavelength of initial intensity and is focussed on the flame cell containing ground state atoms. The atom concentration in the flame cell reduces the initial light intensity. The reduced intensity \( I \) is the measure by the detector. The following equation shows the amount of light absorbed by comparing \( I \) to \( I_0 \): 

\[
A = \log \left( \frac{I}{I_0} \right) = abc
\]

where \( A \) is the absorbance, \( a \) is the absorption coefficient, \( b \) is the length of the light path intercepted by the absorption cell and \( c \) is the concentration of the absorbing species in the absorption cell. This expression defines the absorption \( A \) and is a mathematical statement of Beer's law (Beaty, 1978; Ewing, 1990).

For measuring total and available iron and zinc the SpectrAA OS/2 Varian was used. This SpectrAA system offered extensive analytical capability and a range of functions designed to help set up and easily run even the most complicated atomic absorption determination. The Varian SpectrAA instrument interface runs under IBM's Operating System/2 (OS/2). Varian SpectrAA (Flame) has been carefully designed, so that when used properly it is an accurate, fast, flexible and safe analytical system (Welz and Sperling, 1999).

The burner used in this experiment was an air-acetylene burner which includes an interlock spigot designed to inhibit ignition if either a burner is not fitted to the spray chamber, or a burner is fitted which is not suitable for the flame gas mixture selected.
4.1.3.1 Removing and cleaning the burner

To obtain accurate results the burner must be clean before and after each day of use. For cleaning the burner a clean business card with the Varian burner cleaning and alignment strip on both sides with a brass polish was used. The burner was scrubbed with a soft nylon brush using hot water and detergent. An ultrasonic bath containing a dilute solution of a non-ionic detergent was used, and subsequently the burner was rinsed with hot running water and followed by distilled water. Finally, a dry white business card was used to remove water from inside the burner slot.

4.1.3.2 Removing and cleaning the spray chamber

Due to the use of organic solutions in this experiment, cleaning the spray chamber before and after every analysis was necessary. Before and after every set of analyses the spray chamber was removed, dismantled and aspirated with a weak (approximately 0.1%) solution of a non-ionic detergent for 10 minutes. All parts were thoroughly rinsed with hot water, distilled water and then dried completely.
4.1.4 pH meter

The measurement of pH was important. A (Orion, model 3001) pH meter was used to measure and for adjusting the pH of the pepsin solution.

4.1.5 Shaking water bath

A Tecator 1024 shaking water bath was used. The shaking frequency was adjusted for the first two hours (60) and then 40 for the second two hour (Goni et al., 1996). The unit permits adjustment of the shaking frequency in the range of 10 - 170 strokes per minute.

4.2 Methods

4.2.1 Measurements:

Chemical measurements were determined on triplicate aliquots of each sample described in AOAC (1990).
4.2.2 Preparation of samples, chemicals and glassware:

All glassware used in this study were washed and soaked in nitric acid (1:1) for 30 minutes (AOAC, 1990). They were then rinsed thoroughly with tap water and subsequently with distilled water and left to air dry on a plastic rack.

4.2.2.1 Sample preparation

Samples were weighed into ten grams lots of whole rice and soaked in 100 ml beakers. Half of the sample was soaked in plain water and the other half was soaked in salted water (1:5 W/V). All samples were soaked for the period of 2, 4, 8, 12 and 24 hours at room temperature (~25 °C). The samples were then drained and placed in numbered dishes and put into a cool muffle furnace (less than 250 °C), and ashed at 525 °C overnight.

4.2.3 Moisture determination air oven method

This method involves the measurement of the weight lost due to evaporation of water. With cereal, only a proportion of the water present (free water) is lost at the drying temperature of 105°C. The remainder (bound water) is difficult to remove and appears to be associated with protein. The proportion of free water lost increases
as the temperature is increased, therefore, it is important to compare the results obtained using the identical conditions of drying.

4.2.4 Total iron and zinc measurements

Total iron and zinc content of rice and its various treatments were measured by flame atomic absorption spectroscopy (AAS, 800) before and after soaking in plain and salted water (1%) for the period of 2, 4, 8, 12, and 24 hours at room temperature.

In the second part of the study, samples of rice were cooked before and after soaking in plain and salted water with two different cooking methods (absorption and boil methods).

Subsequently, both the soaked and cooked rice samples were analysed for mineral (Zn and Fe) and phytate content according to the AOAC (1990) and BRI, 1997 methods respectively.
4.2.5 Cooking methods:

4.2.5.1 Absorption method

Thirty ml of water was boiled in a 100 ml beaker and 10 grams of soaked rice added, stirred, the heat lowered and the rice covered with a watch glass, and let stand till all the water was absorbed.

4.2.5.2 Boiling method

One hundred ml of water was boiled, then 10 grams of pre soaked rice was added to the boiled water, stirred, boiled for 15 minute and drained. The resultant rice sample was steamed for 5 minutes.

4.2.6 Phytate measurements

Dry powdered samples were used for phytate analyses. The weighed (10g) soaked and cooked rice were dried in an airflow oven. They were then ground for 20 second in a grinder (Tecator, Knifetec 1095) containing a stainless steel blade and passed through 2.5mm plastic mesh. All samples were extracted with 40 ml of 0.38M HCl / 10% Na₂SO₄ for 3 hours using a magnetic stirrer (BRI, 1997). The complete method is described in appendix.

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4.2.7 Iron and zinc bioavailability

4.2.7.1 Preparation of samples

In the first part of the study, samples were soaked in plain and salted water. The soaked samples were then dried at 40 °C (Pushpanjali and Fenwick, 1994) in an air flow oven for 3-4 hours. The dried samples were ground for 20 seconds in a grinder (Tecator, Knifetec 1095) containing a stainless steel blade and passed through 2.5 mm plastic mesh. This powder was used for determining the available iron and zinc by the in vitro method as described by Khokhar and khokhar (1996). This method is based on the release of ionisable iron from foods subjected to treatment with pepsin-HCL at pH = 1.35 and subsequent adjustment of pH to 7.5, simulating somewhat the conditions prevailing in the stomach and intestine respectively. The validity of this procedure has been evaluated (Kies, 1982).

In the second part of this experiment un-soaked and soaked rice was cooked using different cooking methods (boil method and absorption method). The cooked samples were dried at 40 °C in an air flow oven overnight. They were then ground for 20 seconds in the same grinder that was used for uncooked rice and passed through 2.5 mm plastic mesh. The powder was used for determining available iron and zinc by the in vitro method as described by AOAC (1990).
4.2.8 Statistical analysis

Standard statistical methods were used for calculating mean values and standard errors of mean (s.e.m). Mean values were compared using the student’s t-test. All the experiments were conducted in triplicate and the results obtained were the means of triplicate concurrent values. Data were analysed by the use of computer program Excel 5 and Duncan's multiple range test (Costate) at the 5% level of the probability.
4.2.9 Experimental design

Study one.

Study two.

Study three.
4.3 RESULTS AND DISCUSSION

4.3.1 Total iron, zinc and phytate content of Basmati rice

Rice is the staple food for hundreds of million of people (Tuntawiroon et al., 1990) and may supply up to 80% of the body’s energy requirements (Hutton, 1999). Although rice appears to contain sufficient iron and zinc (Hallberg, 1974), iron deficiency is common in most developing countries especially in those where rice is the main staple food. There has been an increased awareness of the negative effects of iron deficiency on health and well being in recent years. Several studies reported that the main cause of iron deficiency is nutritional, which means that the diet can not cover the physiological iron requirements. The amount of total iron, total zinc and total phytate in raw Basmati rice with a moisture content of 12% were measured (Table 14).

<table>
<thead>
<tr>
<th></th>
<th>(mg/100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73 ± 0.03</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45 ± 0.09</td>
</tr>
<tr>
<td>Phytate</td>
<td>49.70 ± 2.9</td>
</tr>
</tbody>
</table>

Table 14: Total iron, zinc, and phytate content of unsoaked and uncooked Basmati rice (mg/100g).

Values are average of three replicates ±SD.
4.3.2 Absorption of iron and zinc in Basmati rice

The absorption of iron and zinc from the diet is determined by the dietary intake of these minerals (total iron or total zinc) and the balance between those factors in the diet enhancing absorption and those inhibiting absorption. Phytate is known to form insoluble salts with iron and zinc in the digestive system rendering them unavailable to the body (Dickerson et al., 1984).

Tuntawiroon et al. (1990) reported that the phytate content of rice is much higher than that of wheat. Hallberg et al. (1989) proposed that even small amounts of phytate strongly inhibited the absorption of most minerals including iron. Tuntawiroon et al. (1990) also reported that rice starch had no inhibitory effect on iron absorption and that the inhibition of iron absorption was due to the phytate content of rice.

4.3.3 Soaking of Basmati rice

Soaking was the first step, followed by two different cooking methods to reduce the phytate and increase the bioavailability of iron and zinc. The amount of phytate in rice varies depending upon several factors including the milling technique employed. A well-polished rice has a considerably lower phytate content than unpolished rice (Tuntawiroon et al., 1990). Soaking of rice in plain and salted water considerably reduced the amounts of phytate in Basmati rice (Figure 15). Rosalind et
al., (1998) stated that soaking could be used as practical, non-enzymic household method to reduce the water-soluble form of phytate (sodium or potassium phytate) content of some cereals. The decrease in the phytate contents of Basmati rice as a result of soaking was the highest when it was soaked in salted water for a period of 8 hours (53%). Srivastava and Khokhar (1996) also reported that the losses of phytate were greater when grain legumes were soaked in alkaline solution rather than in plain water. It has been suggested that loss of phytate during soaking might be due to leaching out of the phytate ions into the soaking water under the influence of the concentration gradient (Ene-Obong and Obizoba, 1996). Khokhar and Chauhan, (1986) also proposed that soaking of moth bean in plain or salted water was an effective method of lowering phytate content.

Soaking is an integral part of the traditional methods for pre cooking polished rice in Middle Eastern countries. The most important objective of the soaking process is to increase the moisture content, to a level and distribution (within the kernel!) whereby the subsequent cook stage will ensure the complete and uniform cooking of the starch in the rice kernels. Soaking offers the dual advantages of shortening the cooking time as well as rendering the grains nutritionally superior by removing significant amounts of phytate.
Figure 15: The amount of phytate in Basmati rice after soaking in plain or salted water.

Values are average of three replicates ± SD
Soaking of Basmati rice in plain and salted water also significantly (p < 0.01) reduced the amount of total iron (Figure 16) and total zinc (Figure 17) in varying proportions. Total iron and zinc loss increased steadily as soaking time increased. Ene-Obong and Obizoba, (1996) proposed that soaking of the African Yambean for 12 and 24 hours significantly reduced iron content by 29 and 35% respectively. The loss of total iron and total zinc from rice during soaking could be attributed to the leaching of these ions into the soaking medium through simple diffusion. Soaking in salted water seems to remove more iron and zinc up to (46%) and (27%) respectively. Since a salt water solution is known to alter the permeability of the seed coat (Khokhar and Chauhan, 1986) a comparatively greater loss of iron and zinc occurred during soaking in salted water.

Although soaking reduced both total iron and zinc, the latter was more stable during soaking. Ene-Obong and Obizoba, (1996) also stated that zinc is more stable during soaking. Total iron and zinc decreased by 27% and 8% respectively after 4 hour of soaking in plain water (Figure16 and 17).
Figure 16: Total iron in Basmati rice after soaking in plain and salted water.

Values are average of three replicates ±SD.

Figure 17: Total zinc in Basmati rice after soaking in plain and salted water.

Values are average of three replicates ±SD.
The 2, 4, 8, 12 and 24 hr period of soaking were selected because rice is normally soaked 2 to 4 hours prior to cooking or 8 to 12 hour prior to cooking depending upon the variety and age. Old rice eg. 6 months to 1 year old are soaked longer eg 8 to 12 hours. Due to changes in the cooking properties of rice grain during storage the older rice needs to be soaked or cooked longer. Lorenz and Kulp (1991) proposed that change in the cooking properties of rice grains during storage is mostly due to the change in structure-maintaining properties such as endosperm cell walls and endosperm proteins rather than change in the rice starch.

Soaking also resulted in a decrease in the total content of ash (Table 15). This reduction in total ash in Basmati rice could be attributed to the loss of water-soluble ash during soaking. This result was similar to that reported by Bakr (1996).

Table 15: Percent ash in Basmati rice after soaking.

<table>
<thead>
<tr>
<th>Soaking time (Hours)</th>
<th>% ash (Soaked in plain water)</th>
<th>% ash (Soaked in salted water)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>0.4</td>
<td>0.5</td>
</tr>
<tr>
<td>4</td>
<td>0.3</td>
<td>0.5</td>
</tr>
<tr>
<td>8</td>
<td>0.3</td>
<td>0.5</td>
</tr>
<tr>
<td>12</td>
<td>0.3</td>
<td>0.5</td>
</tr>
<tr>
<td>24</td>
<td>0.2</td>
<td>0.3</td>
</tr>
</tbody>
</table>

Values are average of three replicates ±SD.
4.3.4 Molar ratio

The phytate : zinc molar ratio is an important factor for the bioavailability of dietary zinc specially in cases where daily dietary zinc intake is well below the RDI or where the metabolic requirement for zinc is increased such as in growing infants, children, pregnant and lactating women. It is also important in those consuming large amounts of grain and legumes with little or no meat (Morris and Ellis, 1981; Oberleas and Harland, 1981). The molar ratio of phytate : iron and phytate : zinc was 2.6 and 3.6 respectively in Basmati rice. This molar ratio of phytate: zinc decreased to 2.1 after 4 hours of soaking in salted water (Table 16) indicating that more phytate lost than zinc which increased the bioavailabilities of iron and zinc.
Table 16: The phytate: zinc ratio of Basmati rice after different treatments.

<table>
<thead>
<tr>
<th>Soaking time (hour)</th>
<th>0</th>
<th>2</th>
<th>4</th>
<th>8</th>
<th>12</th>
<th>24</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>3.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Soaked in plane water</td>
<td></td>
<td>3.3</td>
<td>2.6</td>
<td>2.7</td>
<td>3.1</td>
<td>3.3</td>
</tr>
<tr>
<td>Soaked in salted water</td>
<td></td>
<td>2.8</td>
<td>2.1</td>
<td>2.0</td>
<td>2.3</td>
<td>2.5</td>
</tr>
<tr>
<td>Cooked (absorption method)a</td>
<td>3.6</td>
<td>3.3</td>
<td>2.7</td>
<td>2.6</td>
<td>3.0</td>
<td>3.4</td>
</tr>
<tr>
<td>Cooked (absorption method)b</td>
<td></td>
<td>3.0</td>
<td>2.1</td>
<td>2.1</td>
<td>2.3</td>
<td>2.4</td>
</tr>
<tr>
<td>Cooked (boil method) a</td>
<td>3.6</td>
<td>3.3</td>
<td>3.6</td>
<td>3.8</td>
<td>3.9</td>
<td>3.9</td>
</tr>
<tr>
<td>Cooked (boil method) b</td>
<td></td>
<td>3.3</td>
<td>2.5</td>
<td>2.4</td>
<td>2.7</td>
<td>2.8</td>
</tr>
</tbody>
</table>

Values are average of three replicates ±SD.
a: soaked in water
b: soaked in salted water

The molar ratio of phytate: iron decreased from 2.6 to 2.0 when Basmati rice was soaked for four hours, in salted water and cooked via the absorption method (Table 17).
Table 17: The phytate: iron ratio of Basmati rice after different treatments.

<table>
<thead>
<tr>
<th>Soaking time (hour)</th>
<th>0</th>
<th>2</th>
<th>4</th>
<th>8</th>
<th>12</th>
<th>24</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>2.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Soaked in plane water</td>
<td>2.5</td>
<td>2.4</td>
<td>2.5</td>
<td>2.9</td>
<td>2.9</td>
<td></td>
</tr>
<tr>
<td>Soaked in salted water</td>
<td>3.0</td>
<td>2.3</td>
<td>2.2</td>
<td>2.4</td>
<td>2.4</td>
<td></td>
</tr>
<tr>
<td>Cooked (absorption method) a</td>
<td>2.6</td>
<td>2.5</td>
<td>2.4</td>
<td>3.0</td>
<td>2.7</td>
<td>2.7</td>
</tr>
<tr>
<td>Cooked (absorption method)b</td>
<td>2.7</td>
<td>2.0</td>
<td>2.1</td>
<td>2.5</td>
<td>2.5</td>
<td></td>
</tr>
<tr>
<td>Cooked (boil method) a</td>
<td>2.7</td>
<td>3.0</td>
<td>2.7</td>
<td>3.1</td>
<td>3.4</td>
<td>3.5</td>
</tr>
<tr>
<td>Cooked (boil method) b</td>
<td>3.9</td>
<td>2.9</td>
<td>2.6</td>
<td>2.9</td>
<td>2.8</td>
<td></td>
</tr>
</tbody>
</table>

Values are average of three replicates ±SD.

a: soaked in water
b: soaked in salted water
4.3.5 Cooking of rice:

Changes resulting from cooking are complex. In the case of cooking rice via the absorption method, rice is cooked substantially on its own, therefore the difference between raw rice and cooked rice reflect only the changes due to cooking. However in the case of the boiling method, the difference between the raw rice and the final cooked rice reflect not only the changes due to cooking but also due to dilution as well as interaction with other ingredients. In the traditional method of rice cooking in Iran with the pre-treatments such as washing and soaking in plain or salted water, many nutrients including minerals such as iron and zinc, vitamins and soluble fibre as well as phytate may be lost from the primary product.

Cooking via the absorption method does not have any affect on the total mineral content of rice, whereas cooking with the boiling method significantly (P< 0.01) reduced the total iron and zinc (Table 18). The phytate content of the Basmati rice also decreased after cooking (boil method) but there was no significant increase on the minerals bioavailability (Table 18). Cooking via the boiling method did decrease iron and zinc, 14 and 11% respectively.
Table 18: The amount of total iron and total zinc in Basmati rice after cooking (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Raw rice</th>
<th>Cooked (boil method)</th>
<th>Cooked (absorption method)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total iron</strong></td>
<td>1.73</td>
<td>1.48</td>
<td>1.69</td>
</tr>
<tr>
<td><strong>Total zinc</strong></td>
<td>1.45</td>
<td>1.29</td>
<td>1.41</td>
</tr>
<tr>
<td><strong>Bioavailable Fe</strong></td>
<td>0.22</td>
<td>0.20</td>
<td>0.21</td>
</tr>
<tr>
<td><strong>Bioavailable Zn</strong></td>
<td>0.25</td>
<td>0.23</td>
<td>0.24</td>
</tr>
<tr>
<td><strong>Phytate</strong></td>
<td>49.7</td>
<td>44.2</td>
<td>48.9</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

The major change in the amount of total iron (Figure 18) and total zinc (Figure 19) as well as the amount of phytate (Figure 20) were observed when the rice was soaked in salted water followed by cooking via the boiling method. Cooking losses vary according to the amount of water used and are greater when excess water is present.
Figure 18: Total iron in Basmati rice after soaking and cooking.

![Graph showing total iron in Basmati rice](image)

Values are average of three replicates ±SD.

a: Soaked in water.
b: Soaked in salted water.

Figure 19: Total zinc in Basmati rice after soaking and cooking.

![Graph showing total zinc in Basmati rice](image)

Values are average of three replicates ±SD.

a: Soaked in water.
b: Soaked in salted water.
Figure 20: Amounts of phytate in Basmati rice after different soaking and cooking.

Values are average of three replicates ±SD.
- a: Soaked in water.
- b: Soaked in salted water.
4.3.6 Phytate measurement:

The phytate content of the rice not only affects iron absorption from the rice but also the iron absorption from other foods in the same meal. Tuntawiroon et al., (1990) reported a threefold difference in absorption between meals served with polished (less phytate) and unpolished (more phytate) rice. Davies (1982) proposed that approximately 70% of the phytate intake of the diet comes from cereals, 20% from fleshy fruits and the remainder from vegetables and nuts. Change in the amount of phytate in rice as affected by domestic pre-treatments is illustrated in Table 17.

Phytate content of the Basmati rice was measured after different periods of soaking, in plain and salted water and two different cooking methods. The total phytate content of whole Basmati rice was 49.7 mg / 100g (Table 18). Soaking of Basmati rice in plain water led to a significantly decreased phytate content (p< 0.01).

Similar observation have been made by Bakr (1996) for phytate in fava bean. A further reduction in phytate content was also noticed after soaking of Basmati rice in salted water which could be due to the leaching out of the phytate ions into the soaking water under the influence of the concentration gradient (Khokhar and Chauhan 1986; Ene-Obong and Obizoba, 1996). The concentration of salt in the soaking media might affect permeability and porosity of the seed coat and thus enhance the removal of the phytate from the seed to the soaking medium. The absorption of water in seeds could also activate phytase resulting in hydrolysis and hence loss of phytic acid (Deshpande and Cheryan, 1983). Similar observations have been made by Khokar and Chauhan (1986) in moth bean.
Table 19: The amount of phytate in Basmati rice after different treatments (mg /100g).

<table>
<thead>
<tr>
<th>Soaking time (hour)</th>
<th>0</th>
<th>2</th>
<th>4</th>
<th>8</th>
<th>12</th>
<th>24</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>49.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>± 2.9</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Soaked in plain water</td>
<td>42.1</td>
<td>33.0</td>
<td>34.2</td>
<td>39.1</td>
<td>40.1</td>
<td></td>
</tr>
<tr>
<td></td>
<td>± 2.6</td>
<td>± 3.7</td>
<td>± 0.9</td>
<td>± 1.5</td>
<td>± 1.5</td>
<td></td>
</tr>
<tr>
<td>Soaked in salted water</td>
<td>33.3</td>
<td>24.1</td>
<td>23.3</td>
<td>25.8</td>
<td>24.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>± 2.2</td>
<td>± 2.7</td>
<td>± 0.7</td>
<td>± 1.3</td>
<td>± 1.4</td>
<td></td>
</tr>
<tr>
<td>Cooked (absorption method)a</td>
<td>48.9</td>
<td>41.1</td>
<td>33.4</td>
<td>32.3</td>
<td>37.4</td>
<td>39.3</td>
</tr>
<tr>
<td></td>
<td>± 3.7</td>
<td>± 3.2</td>
<td>± 2.7</td>
<td>± 4.5</td>
<td>± 1.6</td>
<td>± 2.5</td>
</tr>
<tr>
<td>Cooked (absorption method) b</td>
<td>34.3</td>
<td>24.4</td>
<td>24.1</td>
<td>25.0</td>
<td>25.3</td>
<td></td>
</tr>
<tr>
<td></td>
<td>± 1.4</td>
<td>± 3.1</td>
<td>± 2.2</td>
<td>± 1.8</td>
<td>± 3.7</td>
<td></td>
</tr>
<tr>
<td>Cooked (boil method) a</td>
<td>44.2</td>
<td>37.7</td>
<td>32.9</td>
<td>35.6</td>
<td>38.9</td>
<td>38.2</td>
</tr>
<tr>
<td></td>
<td>± 1.2</td>
<td>± 0.9</td>
<td>± 1.6</td>
<td>± 1.6</td>
<td>± 1.6</td>
<td>± 2.2</td>
</tr>
<tr>
<td>Cooked (boil method) b</td>
<td>31.8</td>
<td>23.7</td>
<td>22.2</td>
<td>24.3</td>
<td>23.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>± 3.5</td>
<td>± 1.7</td>
<td>± 1.0</td>
<td>± 2.4</td>
<td>± 1.1</td>
<td></td>
</tr>
</tbody>
</table>

Values are average of three replicates ±SD.

a: soaked in water
b: soaked in salted water
The total amount of phytate increased slightly after 8 hours of soaking (Table 19) which could be due to reabsorption as well as the hydrolysis of organic phosphate to inorganic phosphate by phytase. Cooking by the absorption method did not have a significant effect on the amount of phytate but cooking by boiling method decreased the phytate (11%). Cooking via the boiling method followed by soaking Basmati rice in salted water for the period of at least 4 hours had the maximum effect of reducing the phytate content. Similar observations have been made by Mameesh and Mantatomar (1993) when they observed 82% decrease in the concentration of phytic acid after cooking the rice via boiling method.

4.3.7 Bioavailability of iron and zinc

In Basmati or white rice despite the relatively high amount of iron and zinc the bioavailable iron and zinc were only about 12 and 17% of the total iron and total zinc respectively (Table 20). Although the amount of total iron was higher than total zinc, in Basmati rice (Table 14), the percent bioavailable zinc was higher than bioavailable iron in this experiment (Table 20). This low bioavailability of iron and zinc in Basmati rice could be due to the high amounts of phytate (49.7 mg/100g) in this rice.
Table 20: Bioavailable iron and zinc in Basmati rice.

<table>
<thead>
<tr>
<th></th>
<th>Iron</th>
<th>Zinc</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bioavailable (mg/100g)</td>
<td>0.22 ± 0.01</td>
<td>0.25 ± 0.02</td>
</tr>
<tr>
<td>Bioavailable (%)</td>
<td>12.7</td>
<td>17.2</td>
</tr>
</tbody>
</table>

Values are average of three replicates ±SD.

4.3.7.1 Bioavailability of iron and zinc after soaking

The ionisable iron and zinc determined at pH = 7.5 can be used as a valid measure to predict iron and zinc bioavailability from foods in human diets (Narasinga and Prabhavathi, 1978). Narasinga Rao and Prabhavathi (1978) demonstrated that both in-vitro and in-vivo methods produced similar results in the range of 1.36-3.89% absorption. Since iron absorption from cereal based diets lies in this range, ionisable level may be applicable to such diets.

Several studies have reported that the bioavailable iron as measured by this invitro method decreases considerably when inhibitors like phytic acid or tannin are included (Disler et al., 1975; Sandberg et al., 1989; Khokhar and Khokhar, 1996). Although rice appears on the basis of chemical analyses to be a good source of trace minerals such as iron and zinc, such minerals are generally poorly utilised by man and other monogastric animals (Toma and curtis, 1986). The negative correlation coefficients between phytate and iron or zinc availability in this study suggests that phytate has an inhibiting effect on the absorption of these minerals. Khokhar and
Khokhar (1996) also reported that phytate had adverse effects on the availability of these minerals. Studies in humans have shown that phytates (Davies and Nightingale, 1975; James et al., 1997) decrease the absorption of nonheme iron from diets. Also physiochemical and nutritional studies have shown that phytates do effectively chelate non-heme iron and thus reduce its bioavailability (Khokhar and Khokhar, 1996).

In the current study, soaking Basmati rice in salted water significantly (p<0.01) increased the bioavailability of iron as well as the bioavailability of zinc (Figure 21 and Figure 22). Although the amounts of total iron and total zinc decreased significantly (p < 0.05), after soaking (Figure 18 and Figure 19) an increase in the bioavailability of iron and zinc, occurred which may be due to the reduction of the phytate content through soaking. These results confirm that even a small reduction of phytate can result in a modest improvement in iron and zinc absorption. Therefore this increase in bioavailability not only increases the bioavailability of iron and zinc from Basmati rice, but it also could extend to the whole nonhaem iron pool in any meal of which they form a part (Bothwell et al., 1979). Due to the relatively high amounts of phytate in Basmati rice, it would be expected that the bioavailability of iron and zinc might be low in diets in which rice was one of the staple foodstuffs.

The bioavailability of iron and zinc increased up to 17.9% and 23% respectively after 4 hours of soaking Basmati rice in salted water. Soaking Basmati rice for longer periods of time did not further effect the bioavailability of these minerals (Figure 21 and Figure 22). Soaking in plain water also significantly (p < 0.05) increased the bioavailability of iron and zinc in Basmati rice but not to the same extent.
Figure 21: Bioavailable iron in Basmati rice after soaking.

Values are average of three replicates ±SD.

Figure 22: Bioavailable zinc in Basmati rice after soaking.

Values are average of three replicates ±SD.
4.3.7.2 Bioavailability of iron and zinc after different cooking

Cooking via absorption method did not result in any significant change on the bioavailability of iron (Figure 23) and zinc (Figure 25) in Basmati rice but cooking via the boiling method followed by soaking decreased the bioavailability of iron and zinc slightly (Figure 24 and 26).
Figure 23: Bioavailable iron in Basmati rice after soaking and cooking (absorption method).

![Graph showing bioavailable iron in Basmati rice over soaking time.](image)

Values are average of three replicates ±SD.

- a: Soaked in plain water.
- b: Soaked in salted water.

Figure 24: Bioavailable iron in Basmati rice after soaking and cooking (boil method).

![Graph showing bioavailable iron in Basmati rice over soaking time.](image)

Values are average of three replicates ±SD.

- a: Soaked in plain water.
- b: Soaked in salted water.
Figure 25: Bioavailable zinc in Basmati rice after soaking and cooking (absorption method).

Values are average of three replicates ± SD.

a: Soaked in water.
b: Soaked in salted water.

Figure 26: Bioavailable zinc in Basmati rice after soaking and cooking (boil method)

Values are average of three replicates ± SD.

a: Soaked in water.
b: Soaked in salted water.
4.3.8 Summary tables of Basmati rice results

Summary tables comparing the bioavailable iron and zinc from the original and remaining total iron and zinc after soaking Basmati rice in plain or salted water are presented.

Table 21: A summary table comparing percent bioavailable iron from original total and remaining total iron after soaking Basmati rice in plain water.

<table>
<thead>
<tr>
<th>Soaking times bioavailable (Hours)</th>
<th>Total iron mg/100g</th>
<th>Bioavailable iron mg/100g</th>
<th>% bioavailable iron (A)</th>
<th>% bioavailable iron (B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.73</td>
<td>0.22 b</td>
<td>12.7</td>
<td>12.7</td>
</tr>
<tr>
<td>2</td>
<td>1.51</td>
<td>0.24 ab</td>
<td>13.9</td>
<td>15.9</td>
</tr>
<tr>
<td>4</td>
<td>1.26</td>
<td>0.27 a</td>
<td>15.6</td>
<td>21.4</td>
</tr>
<tr>
<td>8</td>
<td>1.22</td>
<td>0.26 ab</td>
<td>15.0</td>
<td>21.3</td>
</tr>
<tr>
<td>12</td>
<td>1.23</td>
<td>0.25 ab</td>
<td>14.4</td>
<td>20.3</td>
</tr>
<tr>
<td>24</td>
<td>1.26</td>
<td>0.25 ab</td>
<td>14.4</td>
<td>19.8</td>
</tr>
</tbody>
</table>

Within the column values followed by same letter are not significantly different at P<0.05.
A: Percent of original total iron.
B: Percent of remaining total iron.

Analysis of data using Duncan’s Multiple Range Test shows that the available iron was significantly (P<0.05) increased from 12.7% up to 21.4% (Table 21) after soaking of Basmati rice for four hours in water. The amount of bioavailable iron increased from 0.22 mg/100g of rice to 0.27 mg/100g.
Table 22: A summary table comparing percent bioavailable iron from original total and remaining total iron after soaking Basmati rice in salted water.

<table>
<thead>
<tr>
<th>Soaking times bioavailable (Hours)</th>
<th>Total iron mg/100g</th>
<th>Bioavailable iron mg/100g</th>
<th>% bioavailable iron (A)</th>
<th>% iron</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.73 a</td>
<td>0.22 c</td>
<td>12.7</td>
<td>12.7</td>
</tr>
<tr>
<td>2</td>
<td>1.00 b</td>
<td>0.26 b</td>
<td>15.0</td>
<td>26.0</td>
</tr>
<tr>
<td>4</td>
<td>0.96 bc</td>
<td>0.31 a</td>
<td>17.9</td>
<td>32.3</td>
</tr>
<tr>
<td>8</td>
<td>0.97 bc</td>
<td>0.30 ab</td>
<td>17.3</td>
<td>30.9</td>
</tr>
<tr>
<td>12</td>
<td>0.99 bc</td>
<td>0.28 ab</td>
<td>16.2</td>
<td>28.3</td>
</tr>
<tr>
<td>24</td>
<td>0.93 c</td>
<td>0.28 ab</td>
<td>16.2</td>
<td>30.1</td>
</tr>
</tbody>
</table>

Within the columns values followed by the same letter are not significantly different at P<0.05.
A: Percent of original total iron.
B: Percent of remaining total iron.

Although soaking in salted water significantly (P<0.05) reduced the amount of total iron (Table 22) a significant (P<0.05) increase in the bioavailable iron occurred up to 32% (Table 22) when the rice was soaked for four hours in salted water.

Although soaking Basmati rice in plain water gave a significant increase in bioavailable iron after four hours of soaking (Table 21), comparing the results obtained from soaking Basmati rice in salted water showed that salted water was more effective in increasing the bioavailable iron. Analysis of data using Duncan's Multiple Range Test show a significant (P<0.05) increase in the amount of bioavailable iron (20%) when rice was soaked for four hours in salted water rather than in plain water.
Soaking in water did not result in a significant change in the amounts of total and bioavailable zinc in Basmati rice (Table 23).

Table 23: A summary table comparing percent bioavailable zinc from original total and remaining total zinc after soaking Basmati rice in plain water.

<table>
<thead>
<tr>
<th>Soaking times bioavailable (Hours) (B)</th>
<th>Total zinc mg/100g</th>
<th>Bioavailable zinc mg/100g</th>
<th>% bioavailable zinc (A)</th>
<th>% zinc</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.45 a</td>
<td>0.25 a</td>
<td>17.2</td>
<td>17.2</td>
</tr>
<tr>
<td>2</td>
<td>1.35 ab</td>
<td>0.27 a</td>
<td>18.6</td>
<td>20.0</td>
</tr>
<tr>
<td>4</td>
<td>1.33 ab</td>
<td>0.29 a</td>
<td>20.0</td>
<td>21.8</td>
</tr>
<tr>
<td>8</td>
<td>1.34 ab</td>
<td>0.29 a</td>
<td>20.0</td>
<td>21.6</td>
</tr>
<tr>
<td>12</td>
<td>1.33 ab</td>
<td>0.27 a</td>
<td>18.6</td>
<td>20.3</td>
</tr>
<tr>
<td>24</td>
<td>1.26 b</td>
<td>0.27 a</td>
<td>18.6</td>
<td>21.4</td>
</tr>
</tbody>
</table>

*Within the column values followed by same letter are not significantly different at P<0.05.*
A: Percent of original total zinc.
B: Percent of remaining total zinc.

Soaking in salt water decreased the amount of total zinc, but the amount of bioavailable zinc significantly (P<0.05) increased (Table 23). The bioavailable zinc increased to 32% after 24 hours of soaking Basmati rice in salted water.
Table 24: A summary table comparing percent bioavailable zinc from original total and remaining total zinc after soaking Basmati rice in salted water.

<table>
<thead>
<tr>
<th>Soaking times (Hours)</th>
<th>Total zinc mg/100g</th>
<th>Bioavailable zinc mg/100g</th>
<th>% bioavailable Zinc (A)</th>
<th>% bioavailable zinc (B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.45 a</td>
<td>0.25 c</td>
<td>17.2</td>
<td>17.2</td>
</tr>
<tr>
<td>2</td>
<td>1.24 b</td>
<td>0.30 b</td>
<td>20.7</td>
<td>24.2</td>
</tr>
<tr>
<td>4</td>
<td>1.20 b</td>
<td>0.34 b</td>
<td>23.4</td>
<td>28.3</td>
</tr>
<tr>
<td>8</td>
<td>1.19 b</td>
<td>0.34 ab</td>
<td>23.4</td>
<td>28.6</td>
</tr>
<tr>
<td>12</td>
<td>1.16 b</td>
<td>0.35 a</td>
<td>24.1</td>
<td>30.2</td>
</tr>
<tr>
<td>24</td>
<td>1.05 c</td>
<td>0.34 ab</td>
<td>23.4</td>
<td>32.4</td>
</tr>
</tbody>
</table>

Within the column values followed by same letter are not significantly different at P<0.05.
A: Percent of original total zinc.
B: Percent of remaining total zinc.

Soaking of Basmati rice for four hours in salt water not only significantly (P<0.05) increased the bioavailable iron, it also significantly (P<0.05) increased the bioavailable zinc (0.34mg/100g). Therefore soaking in salt water was an effective method to increase the bioavailability for both iron and zinc in Basmati rice.

The majority of the Iranian subjects soak rice in salted water (50% see chapter 3, Figure 6) but only for 2 hours (see chapter 3, Figure 7) which is less effective than for 4 hours in increasing the bioavailable iron and zinc.
4.4 CONCLUSION

Rice is a principal food crop for about half of the world population. Iron deficiency is common in all developing countries and is frequent in countries where rice is the main staple food (Demaeyer and Adiels-Tagman, 1985). Rice frequently serves as a secondary source of iron (Robinson et al., 1990). Several studies reported that there is a strong relationship between the phytate content in the meal and the level of iron absorption with even small amounts of phytate strongly inhibiting absorption (Hallberg et al., 1989). Therefore more extensive investigation into the relationship between the diet and mineral deficiencies needs to be undertaken particularly in areas where dietary habits may be at least partially responsible for the problem.

The available iron and zinc of the Basmati rice as measured by in vitro method was shown to increase when phytate was reduced. Beard et al. (1988) with human studies reported that phytate decreased the absorption of iron from diets. Sandberg (1991) also reported that phytate is one of the major inhibiting factors for zinc and iron absorption according to in vitro measurement and human studies. Sharma and Khetarpaul (1997) found it imperative to reduce the level of anti-nutrients including phytate to improve the bioavailability of the minerals in the human system.

Measurements of the total and bioavailable iron and zinc and the phytate content of the Basmati rice after traditional Iranian precooking and cooking methods
CHAPTER 5

MEASUREMENTS OF TOTAL AND BIOAVAILABLE IRON AND ZINC IN SUNBROWN NATURAL CALROSE (BROWN RICE).

5. INTRODUCTION

Brown rice is the least processed form of rice with only the outer hull removed while retaining the bran layer. Its flavour has a nutty character and the colour is light tan. The shelf life of brown rice is very short (3-6 months) and is due to its susceptibility to rancidity. The commercial production, marketing and consumption of brown rice as well as its products (flour, oil and bran) is limited (Marshal and Wadsworth, 1994).

The fibre content of brown rice is approximately 3.5 grams per 100 grams (LaBell, 1994). Brown rice contains more nutrients (minerals and vitamins) than does milled (white) rice and has been claimed to be nutritionally superior to white rice (Kennedy, 1980). It has a higher percentage of all nutrients except carbohydrate. Kennedy (1980) found that the iron content of brown rice is two to three times greater than white rice. Besides being rich in nutrients the bran layers also have hypcholesterolemic properties (Kestin et al., 1990). It is often the choice for health food or products with a healthy image although the nutrient content alone is
insufficient to assess the nutritional value of food. Lorenz and Kulp (1991) proposed that even though the nutrient content of brown rice may be higher than milled rice, it does not necessarily have greater food value. McGee (1997) stated that indigestible carbohydrate complex with some nutrients in whole grain flour and therefore speed their passage out of the system, while the same nutrients in white flour does not suffer such losses. Therefore although it is true that brown rice contains more protein, minerals and vitamins than refined white rice, it is also true that some of these nutrients pass through the digestive tract unabsorbed. In the normal diet this drawback might be described as negligible but for people on marginal diets it can have disastrous consequences.

5.1 Method

The method used for this part of the experiment is similar to the method described in chapter 4.

5.2 RESULTS AND DISCUSSION

5.2.1 Absorption of iron and zinc in Sunbrown Natural Calrose

Total iron and zinc as well as phytate content of Sunbrown Natural Calrose rice were measured by the AOAC method (1990). The amounts of total iron and total zinc
were high in this rice (Table 25). Kennedy (1980) proposed that the iron and phosphorus content of brown rice is two to three times greater than white rice. Inspite of high amounts of iron and zinc in Sunbrown Natural Calrose the bioavailability of iron and zinc is only about 9 and 14 percent respectively. This may be due to the high content of antinutrients, such as phytate (65.14 mg/100g) in this rice.

Table 25: Total iron, zinc, phytate and the bioavailability of iron and zinc in Sunbrown Natural Calrose.

<table>
<thead>
<tr>
<th></th>
<th>Total (mg/100g)</th>
<th>Bioavailable (mg/100g)</th>
<th>Bioavailable %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iron</td>
<td>2.51 ± 0.07</td>
<td>0.23 ± 0.01</td>
<td>9.2</td>
</tr>
<tr>
<td>Zinc</td>
<td>1.50 ± 0.01</td>
<td>0.21 ± 0.01</td>
<td>14.0</td>
</tr>
<tr>
<td>Phytate</td>
<td>65.14 ±</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

5.2.2 Soaking of Sunbrown Natural Calrose

The iron content of raw Sunbrown Natural Calrose (brown rice) is greater (2.51 mg/100g) than those previously reported in other brown rice (1.6 mg/100g; Lorenz et al., 1974) and (2.4 mg/100g; Juliano and Bechtel, 1985). With such a high level of iron it may be expected to produce significant available iron. However due to the very high level of phytate (65.14 mg/100g) in this rice (Table 25) only 9.2% is available for absorption.
Processing can reduce some of the deleterious effects of anti-nutritional factors such as phytate (Chitra et al., 1996). Soaking of Sunbrown Natural Calrose under a variety of conditions and times led to significant (P<0.05) reduction in phytate (33-50%). These losses were greater when the rice was soaked in salted water (Table 26). Srivastava and Khokar (1995) proposed that reduced phytate after soaking in water is due mainly to its leaching into the soaking water. Deshpande and Cheryan (1983) also reported that losses of phytate increased when the ionic concentration of the soaking medium was increased. Soaking in water was also reported to be effective in decreasing the phytate content in sorghum (57% after 16 hours of soaking) and in pearl millet (14.8% after 15 hours of soaking) (Agrawal and Chitnis, 1995; Pawar and Parlakar, 1990). In Sunbrown Natural Calrose soaking in plain water decreased phytate content up to 40% after 8 hours of soaking (Table 26). These results could be an important observation because phytate reduces the bioavailability of iron and zinc from foods (Cook et al., 1997).
Table 26: The effect of soaking on the amounts of phytate in Sunbrown Natural Calrose (mg/100g).

<table>
<thead>
<tr>
<th>Soaking time (hours)</th>
<th>Soaked in plain water</th>
<th>Soaked in salted water</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>65.14 ± 4.6</td>
<td>65.14 ± 4.6</td>
</tr>
<tr>
<td>2</td>
<td>53.26 ± 3.7</td>
<td>45.10 ± 3.4</td>
</tr>
<tr>
<td>4</td>
<td>41.00 ± 3.6</td>
<td>36.12 ± 4.1</td>
</tr>
<tr>
<td>8</td>
<td>39.23 ± 4.4</td>
<td>32.35 ± 3.7</td>
</tr>
<tr>
<td>12</td>
<td>42.12 ± 4.1</td>
<td>38.10 ± 2.1</td>
</tr>
<tr>
<td>24</td>
<td>49.18 ± 4.1</td>
<td>40.22 ± 2.0</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

Soaking of Sunbrown Natural Calrose also significantly (p< 0.05) reduced the amounts of total iron by 32 % and 45% after soaking in plain and salted water, respectively (Figure 27 and 28). On the other hand zinc was very stable during soaking. Ene-Obong and Obizoba (1995) also found that zinc was very stable during soaking. Marshall and Wadsworth (1994) observed that the higher nutritive content of brown rice when compared to white rice is due to the fact that iron, potassium, sodium, phosphorus and riboflavin are high in the bran whereas the zinc content of bran layer is very low (Kennedy, 1980). This may be the reason for leaching of significant amounts of iron and phytate rather than zinc during soaking of Sunbrown Natural Calrose in plain or salted water.
Although soaking reduced the amount of total iron (45%), when Sunbrow Natural Calrose (brown rice) was soaked in salted water for the period of (2-8) hours (Figure 28), a significant (p<0.05) increase in the bioavailability of iron occurred (Figure 29). A reduction in the amount of phytate (50%) was evident after soaking of Sunbrow Natural Calrose in salted water (Table 26). Also the ionizable zinc in this rice increased to 23% after 8 hours of soaking in salted water (Figure 30).
Figure 27: Total iron and zinc in Sunbrown Natural Calrose after soaking in water.

Values are average of three replicates ± SD

Figure 28: Total iron and zinc in Sunbrown Natural Calrose after soaking in salted water.

Values are average of three replicates ± SD
Figure 29: Bioavailable iron after soaking of Sunbrown Natural Calrose.

Values are average of three replicates ± SD.

Figure 30: Bioavailable zinc after soaking of Sunbrown Natural Calrose.

Values are average of three replicates ± SD.
5.2.3 The effect of various cooking on the bioavailability of iron and zinc in Sunbrown Natural Calrose

Cooking via the absorption method did not noticeably change the levels of total iron and zinc (Figure 31 and 32). However cooking via the boiling method significantly ($P < 0.05$) reduced the iron (23.5%) content of Sunbrown Natural Calrose (Figure 33). It has been reported that 1/3 of the minerals are lost upon washing and cooking rice when it is washed and cooked in 8 volumes of water (Malakar and Benerjee, 1959). Edem et al., (1994) also reported reductions in iron content (33.3%) in conophor seed during cooking. Cooking using the boil method also further reduced the iron of presoaked rice up to (29%). Both methods of cooking of pre soaked rice reduced phytate up to 55% (Figure 35 and 36). These finding are in agreement with those results reported by (Belavadi et al., 1984; Srivastava and Khokhar, 1995). A decrease in phytate content following by cooking via absorption method may be due to an increase in protein digestibility of the rice (Chitra et al., 1995). When rice was cooked using the absorption method after 8 hours of soaking in plain water, the bioavailability of zinc of Sunbrown Natural Calrose was significantly ($p < 0.05$) increased to 14% (Figure 37). Cooking via the absorption method after soaking of Sunbrown Natural Calrose in salted water for the period of 4-8 hours increased the bioavailability of zinc to 28% (Figure 39). There was a no significant increase ($p<0.05$) in the bioavailability of zinc when Sunbrown Natural Calrose was cooked by the boiling method after soaking in plain water for 4-8 hours (Figure 40).

The ionisable iron content of raw rice at pH 7.5 was 0.23 mg/100g (Table 23).
Cooking with two different methods (Boil method and absorption method) had no significant ($p< 0.05$) increase on the bioavailability of iron in this rice. The availability of iron in this rice increased significantly when it was cooked using the absorption method after pre-soaking for 8 hours in salted water (Figure 37). There was a significant decrease ($p<0.05$) in the amount of bioavailable iron after cooking by the boiling method after soaking in salted water for 2-8 hours (Figure 38). This decrease of bioavailable iron may be due to the reduction of iron (52%) during soaking (in salted water) and boiling of rice. Cooking via the absorption method after soaking in salted water for 8 hours improved ionisable iron and was also highly effective in removing phytate (55%). Layrisse and Martinez-Torres (1972) proposed that phytate not only affects the iron absorption from the food in which it is present but also the iron absorption from other food in the same meal.
Figure 31: Total iron after cooking absorption method in Sunbrown Natural Calrose.

Values are average of three replicates ± SD.

Figure 32: Total zinc after cooking absorption method in Sunbrown Natural Calrose.

Values are average of three replicates ± SD.
Figure 33: total iron after cooking boil method in Sunbrown Natural Calrose.

Values are average of three replicates ± SD.

Figure 34: Total zinc after cooking boil method in Sunbrown Natural Calrose.

Values are average of three replicates ± SD.
Figure 35: The amounts of phytate after soaking and cooking (absorption method) in Sunbrown Natural Calrose.

![Figure 35 Graph]

Values are average of three replicates ± SD.

Figure 36: The amounts of phytate after soaking and cooking (boil method) in Sunbrown Natural Calrose.

![Figure 36 Graph]

Values are average of three replicates ± SD.
Figure 37: Bioavailable iron after cooking (absorption method) in Sunbrown Natural Calrose.

Values are average of three replicates ± SD.

Figure 38: Bioavailable iron after cooking (boil method) in Sunbrown Natural Calrose.

Values are average of three replicates ± SD.
Figure 39: Bioavailable zinc after cooking (absorption method) in Sunbrown Natural Calrose.

![Graph showing bioavailable zinc over soaking time]

Values are average of three replicates ± SD.

Figure 40: Bioavailable zinc after cooking (boil method) in Sunbrown Natural Calrose.

![Graph showing bioavailable zinc over soaking time]

Values are average of three replicates ± SD.
5.2.4 Summary tables of Sunbrown Natural Calrose results

Summary tables comparing the results as well as the percent of bioavailable iron and zinc from the original and remaining total iron and zinc after soaking Sunbrown Natural Calrose (brown rice) in plain or salted water are presented.

Soaking of Sunbrown Natural Calrose significantly decreased the amounts of total iron but it did not show any significant change in the amount of bioavailable iron. (Table 27).

Table 27: A summary table comparing percent bioavailable iron from original total and remaining total iron after soaking Sunbrown Natural Calrose rice in plain water.

<table>
<thead>
<tr>
<th>Soaking times (Hours)</th>
<th>Total iron mg/100g</th>
<th>Bioavailable iron mg/100g</th>
<th>% bioavailable iron (A)</th>
<th>% bioavailable iron (B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>2.51 a</td>
<td>0.23 a</td>
<td>9.2</td>
<td>9.2</td>
</tr>
<tr>
<td>2</td>
<td>1.80 b</td>
<td>0.24 a</td>
<td>9.6</td>
<td>13.3</td>
</tr>
<tr>
<td>4</td>
<td>1.73 b</td>
<td>0.26 a</td>
<td>10.3</td>
<td>15.0</td>
</tr>
<tr>
<td>8</td>
<td>1.71 b</td>
<td>0.25 a</td>
<td>10.0</td>
<td>14.6</td>
</tr>
<tr>
<td>12</td>
<td>1.74 b</td>
<td>0.25 a</td>
<td>10.0</td>
<td>14.4</td>
</tr>
<tr>
<td>24</td>
<td>1.82 b</td>
<td>0.22 a</td>
<td>8.8</td>
<td>12.1</td>
</tr>
</tbody>
</table>

Within the column values followed by same letter are not significantly different at P<0.05.
A: Percent of original total iron.
B: Percent of remaining total iron.
Analysing the data shows a significant increase (p<0.05) in the amount of bioavailable iron (Table 28) after soaking Sunbrown Natural Calrose in salted water. This increase was from 9% to 20% of the remaining iron after soaking for four hours.

**Table 28:** A summary table comparing percent bioavailable iron from original total and remaining total iron after soaking Sunbrown Natural Calrose rice in salted water.

<table>
<thead>
<tr>
<th>Soaking times (Hours)</th>
<th>Total iron mg/100g</th>
<th>Bioavailable iron mg/100g</th>
<th>% bioavailable iron (A)</th>
<th>% bioavailable iron (B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>2.51 a</td>
<td>0.23 b</td>
<td>9.2</td>
<td>9.2</td>
</tr>
<tr>
<td>2</td>
<td>1.52 b</td>
<td>0.27 a</td>
<td>10.7</td>
<td>17.8</td>
</tr>
<tr>
<td>4</td>
<td>1.37 c</td>
<td>0.28 a</td>
<td>11.1</td>
<td>20.4</td>
</tr>
<tr>
<td>8</td>
<td>1.37 c</td>
<td>0.28 a</td>
<td>11.1</td>
<td>20.4</td>
</tr>
<tr>
<td>12</td>
<td>1.38 c</td>
<td>0.25 ab</td>
<td>10.0</td>
<td>18.1</td>
</tr>
<tr>
<td>24</td>
<td>1.37 c</td>
<td>0.25 ab</td>
<td>10.0</td>
<td>18.2</td>
</tr>
</tbody>
</table>

Within the column values followed by same letter are not significantly different at P<0.05.
A: Percent of original total iron.
B: Percent of remaining total iron.

Soaking Sunbrown Natural Calrose in plain water did not show any significant change in the amounts of bioavailable zinc (Table 29). Also soaking in salted water significantly (p<0.05) increased bioavailable zinc in Sunbrown Natural Calrose (Table 30) the 18% of the remaining total zinc.
Table 29: A summary table comparing percent bioavailable zinc from original total and remaining total zinc after soaking Sunbrown Natural Calrose rice in plain water.

<table>
<thead>
<tr>
<th>Soaking times (Hours)</th>
<th>Total zinc mg/100g</th>
<th>Bioavailable zinc mg/100g</th>
<th>% bioavailable zinc (A)</th>
<th>% bioavailable zinc (B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.50 a</td>
<td>0.21 a</td>
<td>14.0</td>
<td>14.0</td>
</tr>
<tr>
<td>2</td>
<td>1.44 ab</td>
<td>0.20 a</td>
<td>13.3</td>
<td>13.9</td>
</tr>
<tr>
<td>4</td>
<td>1.44 ab</td>
<td>0.22 a</td>
<td>14.7</td>
<td>15.3</td>
</tr>
<tr>
<td>8</td>
<td>1.40 b</td>
<td>0.21 a</td>
<td>14.0</td>
<td>15.0</td>
</tr>
<tr>
<td>12</td>
<td>1.40 b</td>
<td>0.19 a</td>
<td>12.7</td>
<td>13.6</td>
</tr>
<tr>
<td>24</td>
<td>1.39 b</td>
<td>0.18 a</td>
<td>12.0</td>
<td>12.9</td>
</tr>
</tbody>
</table>

*Within the column values followed by same letter are not significantly different at P<0.05.*

A: Percent of original total zinc.
B: Percent of remaining total zinc.

Table 30: A summary table comparing percent of bioavailable zinc from original total and remaining total zinc after soaking Sunbrown Natural Calrose rice in salted water.

<table>
<thead>
<tr>
<th>Soaking times (Hours)</th>
<th>Total zinc mg/100g</th>
<th>Bioavailable zinc mg/100g</th>
<th>% bioavailable zinc (A)</th>
<th>% bioavailable zinc (B)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.50 a</td>
<td>0.21 a</td>
<td>14.0</td>
<td>14.0</td>
</tr>
<tr>
<td>2</td>
<td>1.46 ab</td>
<td>0.22 a</td>
<td>14.7</td>
<td>15.1</td>
</tr>
<tr>
<td>4</td>
<td>1.46 ab</td>
<td>0.24 ab</td>
<td>16.0</td>
<td>16.4</td>
</tr>
<tr>
<td>8</td>
<td>1.44 b</td>
<td>0.26 b</td>
<td>17.4</td>
<td>18.0</td>
</tr>
<tr>
<td>12</td>
<td>1.46 ab</td>
<td>0.21 a</td>
<td>14.0</td>
<td>14.4</td>
</tr>
<tr>
<td>24</td>
<td>1.37 c</td>
<td>0.21 a</td>
<td>14.0</td>
<td>15.3</td>
</tr>
</tbody>
</table>

*Within the column values followed by same letter are not significantly different at P<0.05.*

A: Percent of original total zinc.
B: Percent of remaining total zinc.

Soaking in salt water for 8 hours significantly (P<0.05) increased both bioavailable iron and zinc in Sunbrown Natural Calrose (Table 28 and 30), where as
soaking in plain water did not show any significant increase in either the bioavailable iron or zinc (Table 27 and 29).

Milling is the primary difference between white rice and brown rice. It is in the milling process where brown rice becomes white rice. Milling which is often called whitening, removes the outer bran layer of the rice grain. Milling strips off the bran layer, which contain several nutrients of major importance. Comparing the result obtained in this research shows that although the amount of total iron (2.51 mg/100g in brown rice in comparison to 1.73 mg/100g in white rice) is higher in brown rice, there is not a significant difference in the bioavailable iron (0.22 mg/100g in white rice and 0.23 mg/100g in brown rice). The bioavailability of iron is a function of its chemical form and the presence of phytate which can bind iron in the gastrointestinal tract and make it unavailable for absorption. Soaking in salt water may reduce the total iron, but it is an effective way to increase the percent of bioavailable iron in both Basmati rice and Sunbrown Natural Calrose.
5.3 CONCLUSION

Brown rice contains more iron and zinc than white rice but they are more difficult to absorb from the brown rice. McGee (1997) stated that although unrefined cereals contain more nutrients, it does not automatically mean they are more healthful. Washing and soaking of Sunbrown Natural Calrose (brown rice) either in plain water or salted water for two hours or more considerably reduced the amount of total iron. This reduction of total iron was more when Sunbrown Natural Calrose was soaked in salted water (from 2.51mg/100g to 1.37mg/100g). Zinc was more stable during soaking. The amount of zinc reduced from 1.51mg/100g to 1.37mg/100g. The amount of phytate also was reduced when Sunbrown Natural Calrose was soaked either in plain or salted water. Soaking in salted water was more effective for reducing the phytate content of Sunbrown Natural Calrose (brown rice). The maximum reduction in the amount of phytate was observed when Sunbrown Natural Calrose was soaked in salted water for eight hours, from 65.14 to 32.35 mg/100g.

The bioavailable iron in Sunbrown Natural Calrose increased after soaking. Soaking in salted water proved beneficial although it reduced total iron and zinc from 2.51 to 1.37 and 1.50 to 1.37 mg/100g respectively. It did increase bioavailable iron and zinc from 9.2% to 20.4% and from 14% to 18% respectively yielding given an absorptive amount of 0.28mg/100g and 0.26mg/100g respectively.

Comparing the results obtained after soaking and cooking of Sunbrown Natural Calrose rice via the two different cooking method showed that cooking via the absorption method did not have any major effect on the amount of total iron and total
zinc present while cooking via the boiling method after soaking of rice further reduced iron. The maximum bioavailable iron was observed when Sunbrown Natural Calrose rice was soaked in salted water for eight hours and cooked via the absorption method.

The maximum available zinc was observed when Sunbrown Natural Calrose rice was soaked in salt water for eight hours although the cooking method did not have any significant effect on the amount of bioavailable zinc. Soaking in salt water also reduced cooking time which is an important variable in both household and commercial applications.

Thus overall soaking in salted water for eight hours resulted in a decrease of phytate content for Sunbrown Natural Calrose rice and an increased iron absorption from 9.2% to 20.4% of the remaining iron.
CHAPTER 6

EFFECT OF ADDING FRESH AND FROZEN HERBS AND/OR VEGETABLES TO THE RICE DISHES ON THE BIOAVAILABILITY OF IRON AND ZINC.

6. INTRODUCTION

An inadequate dietary intake of iron is the major cause of the anaemia found in both developed and developing countries. Several researchers (WHO, 1972; Ministry of Agriculture, 1995) believe that an increase in the amount of iron in the diet through fortification would be an obvious course of action. Questions exist as to the availability of iron when consumed with various foods. The absorption of iron may be altered when consumed with different foods. These changes are catalysed by the presence of certain foods or food ingredients (Miller et al., 1981 and Bothwell, 1989) such as the enhancement of iron absorption by the presence of meat or by the presence of vitamin C (Hunt, 1990 and Fairweather-Tait, 1995) and the inhibition of iron absorption by the presence of phytate or oxalate (Brune et al., 1992 and Cook et al., 1997).

Iron absorption may change as a result of certain common food processing operations including washing or thermal processing. Whether or not changes in
availability are the result of change in the iron itself or due to some synergism with the food (Power, 1983) is not known. However if it can be established that improved availability of iron depends upon the chemical environment of the iron in the food itself, food may thus be designed or selected in which the iron is maximally available.

6.1 Materials and methods

6.1.1 Selection and preparation of rice dishes

In order to select the rice dishes that would be reasonably representative of commonly consumed Iranian rice dishes the survey conducted in Chapter 3 was further analysed. The most commonly consumed rice dishes are listed below:

6.1.1.1 Rice with herbs

- Basmati rice and dill
- Basmati rice and parsley
- Basmati rice and coriander
- Basmati rice and chive
6.1.1.2 Rice with vegetables

- Basmati rice and cauliflowers
- Basmati rice and cabbage

6.1.1.3 Rice with legumes

- Basmati rice and green beans
- Basmati rice and lentil
- Basmati rice and broad beans

6.1.2 Chemical measurements

Chemical measurements were conducted on triplicate aliquot's of each rice meal preparation. Total iron and zinc were measured in the different herbs, vegetables and legumes used in the rice dishes by the method described in AOAC (1990) using flame atomic absorption spectroscopy (AAS). All the measurements were repeated with Basmati rice and herbs, vegetables and legumes in triplicate. The in vitro availability of iron and zinc from different rice meal combinations was estimated using the method developed by Rao and Prabhavathi (1978) and used by Pushpanjali and Khokhar (1995). Each rice meal combination was incubated with pepsin- HCl at 37°C at pH = 1.35 for 90 minutes (Pepsin from Sigma 70000). It was
centrifuged, filtered, adjusted to pH = 7.5 and incubated for 45 min. Ionisable iron was estimated in the final filtrate by AAS.

In all rice dishes, 10 gram of Basmati rice were cooked with 2.5 gram of either a herb, a vegetable or a legume. The cooking method used for this study was the absorption method. Total iron and zinc as well as bioavailable iron and zinc were measured separately according to the AOAC method (1990).

Total and bioavailable iron and zinc were also measured separately in all herbs, vegetables and legumes as well as in the rice dishes.

The amount of total iron and zinc as well as bioavailable iron and zinc were then compared with total and bioavailable iron and zinc in plain rice as well as the herbs, vegetables and legumes separately.

The percent change in the total and bioavailable iron and zinc of the rice dish after adding fresh, frozen and dried herbs, vegetables and legumes to the rice dishes was also measured. The equation used to calculate the theoretical percent change in bioavailable iron and zinc in rice meals is as follow:

\[
\% \text{ change in iron and zinc bioavailability} = a_1 \frac{d_4 + a_2 d_2}{100}
\]

where:

\[a_1 = \text{bioavailable iron or zinc in rice}\]
\[ d_1 = \% \text{ rice used} \]

\[ a_2 = \text{bioavailable iron or zinc} \]

\[ d_2 = \% \text{ herbs, vegetables or legumes used} \]

6.1.3 **Experimental design**

- Basmati rice
  - Cooked
    - Absorption method
      - Added
        - Legumes
        - Herbs
        - Vegetables
          - Dried, fresh or frozen
            - Measurements
              - Bioavailable iron
              - Bioavailable zinc
6.2 RESULTS AND DISCUSSION

The enhancing effects of certain fruit or vegetable juices on iron absorption have previously been demonstrated in several studies. Ballot et al. (1987) reported that paw paw could have a significant positive effect on iron nutrition of individuals if it is regularly present in staple cereal diet with low iron availability. Similar finding were reported by Hallberg (1974) when with the addition of a portion of paw paw containing 66 mg ascorbic acid, iron absorption from a maize meal increased from 0.014 to 0.088. In the current study, select herbs, vegetables and legumes in traditional Iranian rice dishes were tested for their ability to modify iron absorption.

6.2.1 Total and bioavailable iron and zinc in herbs used in Iranian rice dishes

Total and bioavailable iron and zinc were measured in the herbs used in rice dishes (Table 31). The results show that the amount of total iron is higher than the total zinc but the percent bioavailable zinc is higher in those herbs. Among herbs coriander has a high zinc content (1.6 mg/100g). The percent bioavailable zinc also was high in coriander (Table 29).
Table 31: Total iron, zinc, and the bioavailability of iron and zinc in herbs (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Dill</th>
<th>Parsley</th>
<th>Coriander</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>4.0</td>
<td>4.8</td>
<td>5.0</td>
</tr>
<tr>
<td>Total Zinc</td>
<td>0.9</td>
<td>1.0</td>
<td>1.6</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.7</td>
<td>0.8</td>
<td>0.8</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.2</td>
<td>0.3</td>
<td>0.6</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>17.5</td>
<td>16.6</td>
<td>16.0</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>22.0</td>
<td>30.0</td>
<td>37.5</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

6.2.2 The absorption of iron and zinc from rice and herb dishes

The iron absorption from Basmati rice was improved slightly when herbs such as coriander, parsley or dill were added. The main cause for this increase is most likely the enhancing effect of the ascorbic acid of the added herbs. Fairweather-Tait et al. (1995) reported two-fold increase in the bioavailability of iron from breakfast cereal and whole meal bread when consumed with a drink containing 50 mg of ascorbic acid. Hallberg and Rossander (1984) also stated that the addition of 50mg ascorbic acid could significantly increase the absorption of non-haem iron in black beans. Total and bioavailable iron as measured in parsley, coriander and dill as well
as in rice dishes containing Basmati rice and parsley, Basmati rice and coriander, and Basmati rice and dill are presented in Table 32, 33 and 34, respectively. Although the overall pattern of the results was similar, coriander was most effective in enhancing the iron absorption from the rice meal.

Table 32: Total and bioavailable iron and zinc in Basmati rice, parsley and a rice dish containing Basmati rice and parsley (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Parsley</th>
<th>Rice + parsley</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>4.8</td>
<td>2.1</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>1.0</td>
<td>1.4</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.8</td>
<td>0.31</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.30</td>
<td>0.30</td>
</tr>
<tr>
<td>% bioavailable iron</td>
<td>12.70</td>
<td>16.60</td>
<td>14.80</td>
</tr>
<tr>
<td>% bioavailable zinc</td>
<td>17.20</td>
<td>30.00</td>
<td>21.40</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
Table 33: Total and bioavailable iron and zinc in Basmati rice, coriander and a rice dish containing Basmati rice and coriander.

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Coriander</th>
<th>Rice + coriander</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>5.0</td>
<td>2.1</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>1.6</td>
<td>1.5</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.8</td>
<td>0.35</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.6</td>
<td>0.29</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>16</td>
<td>16.7</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.2</td>
<td>37.5</td>
<td>17.3</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

Table 34: Total and bioavailable iron and zinc in Basmati rice, dill and a rice dish containing Basmati rice and dill.

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Dill</th>
<th>Rice + dill</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>4.0</td>
<td>2.0</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>0.9</td>
<td>1.4</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.7</td>
<td>0.30</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.2</td>
<td>0.28</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>17.5</td>
<td>15.0</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.2</td>
<td>22.0</td>
<td>17.1</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
The amount of ascorbic acid in the three herbs (dill, coriander and parsley) were compared with parsley having the highest amount (Table 3, see page 63). Although parsley contained the most ascorbic acid (190mg/100g) the absorption of iron from the meal with parsley and Basmati rice increased in bioavailability by only 2% (Table 32). It is probable that this limited increase was due to the oxalic content of the parsley (Table 2, see page 46). Ballot et al. (1987) stated that addition of 1 gram calcium oxalate to cabbage reduced iron absorption by 39%.

6.2.3 Bioavailable iron and zinc after soaking and cooking in rice dishes

As soaking in salted water and cooking via the absorption method was the most appropriate method of soaking and cooking to obtain maximum bioavailable iron and zinc, the rice dishes were treated in a similar manner and bioavailable iron and zinc were measured. The amounts of bioavailable iron and zinc are shown in Table (35 and 36).
Table 35: Bioavailable iron in rice dishes containing Basmati rice and herbs after soaking in salted water and cooking via the absorption method.

<table>
<thead>
<tr>
<th>Soaking time (hr)</th>
<th>Plain rice</th>
<th>Rice + parsley</th>
<th>Rice + dill</th>
<th>Rice + coriander</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.22</td>
<td>0.32</td>
<td>0.30</td>
<td>0.35</td>
</tr>
<tr>
<td>2</td>
<td>0.27</td>
<td>0.37</td>
<td>0.36</td>
<td>0.38</td>
</tr>
<tr>
<td>4</td>
<td>0.32</td>
<td>0.39</td>
<td>0.40</td>
<td>0.41</td>
</tr>
<tr>
<td>8</td>
<td>0.32</td>
<td>0.41</td>
<td>0.41</td>
<td>0.40</td>
</tr>
<tr>
<td>12</td>
<td>0.30</td>
<td>0.40</td>
<td>0.39</td>
<td>0.40</td>
</tr>
<tr>
<td>24</td>
<td>0.29</td>
<td>0.40</td>
<td>0.39</td>
<td>0.41</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

The results confirm that in unsoaked rice the phytic acid content of rice is not a problem where herbs containing ascorbic acid are added. Vitamin C content of herbs used in the rice dishes increased markedly the amount of iron absorbed from these rice dishes. It has also been observed that when rice is soaked in salted water and cooked with herbs via the absorption method more iron is absorbed (Table 35).
Table 36: Bioavailable zinc in rice dishes containing Basmati rice and herbs after soaking in salted water and cooking via absorption method.

<table>
<thead>
<tr>
<th></th>
<th>Plain rice</th>
<th>Rice + parsley</th>
<th>Rice + dill</th>
<th>Rice + coriander</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.25</td>
<td>0.30</td>
<td>0.28</td>
<td>0.29</td>
</tr>
<tr>
<td>2</td>
<td>0.31</td>
<td>0.31</td>
<td>0.29</td>
<td>0.37</td>
</tr>
<tr>
<td>4</td>
<td>0.33</td>
<td>0.32</td>
<td>0.29</td>
<td>0.38</td>
</tr>
<tr>
<td>8</td>
<td>0.35</td>
<td>0.34</td>
<td>0.32</td>
<td>0.39</td>
</tr>
<tr>
<td>12</td>
<td>0.35</td>
<td>0.33</td>
<td>0.30</td>
<td>0.40</td>
</tr>
<tr>
<td>24</td>
<td>0.34</td>
<td>0.33</td>
<td>0.31</td>
<td>0.39</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

The bioavailable zinc also were measured in rice meals containing herbs such as parsley, dill or coriander. Among the herbs parsley was the most effective herb to increase the bioavailable zinc in rice dish without soaking the rice. As parsley contain huge amounts of vitamin C (190 mg/100g) in compare with the other herbs (see Table 3 page 63) this could be the reason. Results indicated that both, soaking or adding herbs could increase the bioavailable zinc in rice dishes.
6.2.4 Total and bioavailable iron and zinc in vegetables used in Iranian rice dishes

Table 37 shows the amount of total and the bioavailable iron and zinc in vegetables. Cauliflower had the highest content of iron (1.6 mg/100g) and zinc (0.44) in comparison with savoy cabbage and carrot. The amount of total iron in cauliflower is very close to the total iron content of Basmati rice (1.73mg/100g) whereas the bioavailable iron in cauliflower (0.37mg/100g) was higher than bioavailable iron in Basmati rice (0.22mg/100g).

Table 37: Total iron, zinc, and the bioavailability of iron and zinc in selected vegetables (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Cauliflower</th>
<th>Savoy cabbage</th>
<th>Carrot</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.63</td>
<td>1.36</td>
<td>0.50</td>
</tr>
<tr>
<td>Total Zinc</td>
<td>0.44</td>
<td>0.29</td>
<td>0.36</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.37</td>
<td>0.35</td>
<td>0.20</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.18</td>
<td>0.08</td>
<td>0.15</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>22.6</td>
<td>25.7</td>
<td>40.0</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>41.9</td>
<td>27.6</td>
<td>41.6</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
6.2.5 The bioavailable iron and zinc from rice and vegetable dishes

The bioavailable iron was also measured in dishes containing Basmati rice and a vegetable. The bioavailable iron significantly (p<0.05) increased in a dish containing Basmati rice and cauliflower (Table 38). This increase could be due to the effects of the vitamin C content of cauliflower. Hallberg (1981) reported that the addition of cauliflower containing approximately 70 mg of ascorbic acid to a vegetarian meal trebled the amount of iron absorption. Cauliflower and savoy cabbage both contain vitamin C (43 and 62mg/100g respectively) and carotene (50 and 995μg/100g) respectively (Holland et al., 1992). The level of ascorbic acid in the diet has been found to be an important factor in determining nonhaem iron absorption (Cook and Monsen, 1977; Monsen et al., 1978). Gillooly et al. (1983) stated that in diets containing little or no meat, the ascorbic acid content of the components of a meal should be regarded as a crucial factor in determining nonhaem iron absorption.

Garcia-Casal et al. (1998) stated that carotene may form a complex with iron, keeping it soluble in the intestinal lumen and preventing the inhibitory effect of phytates on iron absorption. Kies (1982) stated that to increase non-haem iron absorption both the nonhaem iron and the ascorbic acid must be consumed at the same time. Therefore as plant origin foods are important sources of ascorbic acid, using more plant based foods leads to increased consumption of ascorbic acid which in turn increases the absorption of iron.
Addition of cauliflower or savoy cabbage into Basmati rice enhanced the bioavailable iron from 0.22 mg/100g to 0.31 mg/100g and zinc from 0.25 mg/100g to 0.35 mg/100g in this experiment (Table 38 and 39).

Table 38: Total iron, zinc, and the bioavailability of iron and zinc in Basmati rice, Cauliflower and in a dish containing cauliflower and Basmati rice (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Cauliflower</th>
<th>Cauliflower + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>1.63</td>
<td>1.70</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>0.44</td>
<td>1.25</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.37</td>
<td>0.31</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.18</td>
<td>0.29</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.70</td>
<td>22.60</td>
<td>18.20</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.20</td>
<td>41.90</td>
<td>23.20</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
Table 39: Total iron, zinc, and the bioavailability of iron and zinc in Basmati rice, savoy and in a dish contains savoy cabbage and Basmati rice (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Savoy cabbage</th>
<th>Savoy cabbage + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>1.36</td>
<td>1.65</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>0.29</td>
<td>1.22</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.35</td>
<td>0.31</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.18</td>
<td>0.29</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>25.7</td>
<td>18.8</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.2</td>
<td>27.6</td>
<td>22.1</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

Table 40: Total iron, zinc, and the bioavailability of iron and zinc in Basmati rice, carrot and in a dish containing carrot and Basmati rice (mg/100g).

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Carrot</th>
<th>Carrot + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>0.50</td>
<td>1.48</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>0.36</td>
<td>1.23</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.20</td>
<td>0.29</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.15</td>
<td>0.24</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.70</td>
<td>40.00</td>
<td>19.50</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.20</td>
<td>41.60</td>
<td>16.30</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
The bioavailable iron in a rice dish containing Basmati rice and carrot increased about 7% (Table 40). Carrot contains a large amount of pro vitamin A (WHO, 1988). Roodenberg et al. (1996) stated that vitamin A may improve the utilisation of ingested iron. Bloem et al. (1989) found a direct correlation between plasma level of vitamin A and blood levels of hemoglobin. Garcia-Casal et al. (1998) stated that concentration of β carotene in the diet could prevent the inhibitory effect of phytates on iron absorption and therefore increase the bioavailability more than threefold in the rice dish. β Carotene content of carrot in the rice dish may form a complex with iron, keeping it soluble in the intestinal lumen and therefore preventing the inhibitory effect of phytates on iron absorption. Roodenberg et al. (1996) also suggested that vitamin A could improve the utilisation of ingested iron and effects the iron concentrations.

The comparison between % bioavailable iron and the expected bioavailable iron in rice dishes shows that % bioavailable iron increased in all rice dishes. The maximum and significant (p <0.05) increase was observed when coriander was added to the rice (Figure 41). The ascorbic acid content of coriander may have caused the enhancement of the absorption of iron from that rice dish. Ballot et al. (1987) stated that the ability of various fruit juices to enhance iron absorption from a rice meal is correlated with the ascorbic acid content. Coulthate and Davies (1994) stated that ascorbic acid is shown to enhances iron absorption by converting ferric iron to ferrous whose hydroxide is moderately soluble. Brock (1996) also stated that the amount of iron available for absorption is depends on the composition of that diet which he mentioned the presence of vitamin C and absence of phytate.
The percent bioavailable zinc significantly (p < 0.05) increased when parsley or coriander were added to the Basmati rice (Figure 42). Again, this increase in the bioavailable zinc could be due to the ascorbic acid content of rice dish.

Between the vegetables, cauliflower was the most effective vegetable to increase the bioavailable iron (Figure 43). This significant (p<0.05) increase could be due to the both ascorbic acid and carotene content of the cauliflower (50 mg/100g). Wolters et al. (1993) reported that ascorbic acid could increase the availability of iron. Gillooly et al., (1983) stated that ascorbic acid owes this ability to reduce properties and chelate iron and its promoting influence on iron absorption is dose dependent. Carotene can also be classified as anti-oxidant which can solublise iron of the diet (Margen, 1992). Adding cauliflower or carrot to the Basmati rice not only increased the bioavailable iron, it also significantly (p < 0.05) increased bioavailable zinc in that rice dish (Figure 44).
Figure 41: Comparison of % bioavailable iron with the expected bioavailable iron in rice dishes containing Basmati rice and herbs.

Values are average of three replication ± SD.

Figure 42: Comparison of % bioavailable zinc with the expected bioavailable zinc in rice dishes contain Basmati rice and herbs.

Values are average of three replicates ± SD.
Figure 43: Comparison of % bioavailable iron with the expected bioavailable iron in rice dishes contain Basmati rice and vegetables.

Values are average of three replicates ± SD.

Figure 44: Comparison of % bioavailable zinc with the expected bioavailable zinc in rice dishes contain Basmati rice and vegetables.

Values are average of three replicates ± SD.
6.2.6 Total iron and zinc in legumes

The amounts of total iron and zinc were also measured in legumes used in the Iranian rice dishes. Among legumes lentil had the highest iron content of 4.51 mg/100g and mung bean had the highest zinc content of 2.9 mg/100g (Table 41). Although lentil had a high iron content, the bioavailable iron was only 0.25 mg/100g or 5.5 percent of the total. This low bioavailability could be due to the high phosphate content of lentil (350 mg/100g) (Holland et al., 1991).

Although total iron in the lentil and mung bean were four times greater than the total iron in broad beans and green beans, the bioavailable iron was similar in all. The bioavailable zinc was very high in mung bean (1.19mg/100g) and lentil (1.29mg/100g) (Table 41).

<table>
<thead>
<tr>
<th></th>
<th>Mung bean</th>
<th>Lentil</th>
<th>Broad bean</th>
<th>Green bean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>3.6</td>
<td>4.51</td>
<td>1.2</td>
<td>1</td>
</tr>
<tr>
<td>Total zinc</td>
<td>2.9</td>
<td>2.51</td>
<td>1.1</td>
<td>0.3</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.25</td>
<td>0.25</td>
<td>0.27</td>
<td>0.23</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>1.19</td>
<td>1.29</td>
<td>0.3</td>
<td>0.06</td>
</tr>
<tr>
<td>% bioavailable iron</td>
<td>6.9</td>
<td>5.5</td>
<td>22.5</td>
<td>23</td>
</tr>
<tr>
<td>% bioavailable zinc</td>
<td>41</td>
<td>51</td>
<td>27.3</td>
<td>20</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
6.2.7 Total and bioavailable iron and zinc in the rice dishes containing Basmati rice and legumes

Total and bioavailable iron and zinc also were measured in the rice dishes such as Basmati rice and lentil, Basmati rice and mung bean, Basmati rice and broad bean, and Basmati rice and green beans (Table 42). The highest bioavailable iron was observed in the rice dishes contain Basmati rice and lentil (0.40 mg/100g) (Table 33). Although lentil had a very low bioavailable iron of only 5.5%, and Basmati rice had a bioavailable iron of only 12.7%, the bioavailability of iron significantly (p < 0.05) increased up to 17.5% in the rice dish contain Basmati and lentil. The bioavailable zinc also increased significantly (p < 0.05) and approximately doubled in the rice dish contain Basmati rice and mung bean as well as Basmati rice and lentil (Table 43). This increase in the bioavailable iron and zinc may be is due to the high riboflavin (0.27 mg/100g) (Table 3 see chapter 2) content of the lentil. Agte et al. (1998) stated that riboflavin has the capacity to form complexes and supplementation of riboflavin may result in increased absorption of zinc and iron.
Table 42: Total and bioavailable iron and zinc in the Basmati rice, green bean as well as Iranian rice dish contain Basmati rice and green bean.

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Green bean</th>
<th>Green bean + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>1</td>
<td>1.58</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>0.3</td>
<td>1.22</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.23</td>
<td>0.27</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.06</td>
<td>0.26</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>23</td>
<td>17</td>
</tr>
<tr>
<td>% of bioavailable zn</td>
<td>17.2</td>
<td>20</td>
<td>21.3</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
Table 43: Total and bioavailable iron and zinc in the Basmati rice, lentil as well as Iranian rice dish contain Basmati rice and lentil.

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Lentil</th>
<th>Lentil + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>4.51</td>
<td>2.28</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>2.51</td>
<td>1.66</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.25</td>
<td>0.40</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>1.29</td>
<td>0.56</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>5.5</td>
<td>17.5</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.2</td>
<td>51</td>
<td>33.7</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

The bioavailable iron and zinc in the rice dish contain Basmati and broad bean as well as Basmati rice and green bean was 5% more than plain rice (Table 42 and 44). Although the amount of total zinc in green bean was only 0.3mg/100g (Table 42) in compare with broad bean 1.1 mg/100g (Table 44), The percent of bioavailable zinc in both rice dishes (Basmati and broad bean (22.5mg/100g) and Basmati and green bean (21.3mg/100g) was similar (Table 44 and 42 respectively). This increase in the bioavailable iron and zinc could be due to the vitamin C as well as carotene content of broad bean (8 and 0.22mg/100g) (Table 3 page 63).
Table 44: Total and bioavailable iron and zinc in the Basmati rice, broad bean as well as Iranian rice dish contain Basmati rice and broad bean.

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Broad bean</th>
<th>Broad bean + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>1.2</td>
<td>1.62</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>1.1</td>
<td>1.38</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.27</td>
<td>0.29</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>0.3</td>
<td>0.31</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>22.5</td>
<td>17.9</td>
</tr>
<tr>
<td>% of bioavailable zinc</td>
<td>17.2</td>
<td>27.3</td>
<td>22.5</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.
Table 45: Total and bioavailable iron and zinc in the Basmati rice, mung bean as well as Iranian rice dish contain Basmati rice and mung bean.

<table>
<thead>
<tr>
<th></th>
<th>Basmati rice</th>
<th>Mung bean</th>
<th>Mung bean + rice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total iron</td>
<td>1.73</td>
<td>3.6</td>
<td>2.10</td>
</tr>
<tr>
<td>Total zinc</td>
<td>1.45</td>
<td>2.9</td>
<td>1.74</td>
</tr>
<tr>
<td>Bioavailable iron</td>
<td>0.22</td>
<td>0.25</td>
<td>0.29</td>
</tr>
<tr>
<td>Bioavailable zinc</td>
<td>0.25</td>
<td>1.19</td>
<td>0.56</td>
</tr>
<tr>
<td>% of bioavailable iron</td>
<td>12.7</td>
<td>6.9</td>
<td>13.8</td>
</tr>
<tr>
<td>% of bioavailable zn</td>
<td>17.2</td>
<td>41</td>
<td>32.2</td>
</tr>
</tbody>
</table>

Values are average of three replicates ± SD.

6.2.8 Expected bioavailable iron and zinc in the Iranian rice dishes

Adding lentil to the Basmati rice significantly increased the percent bioavailable iron and zinc (Figure 45 and 46). A comparison between the expected and real bioavailable iron and zinc in the different rice dishes shows that bioavailable zinc were significantly (p< 0.05) increased in the rice dishes including Basmati rice and mung bean, Basmati rice and lentil (Figure 46), whereas bioavailable iron significantly increased in rice dishes contain Basmati rice lentil, Basmati rice and broad bean as well as Basmati rice and Green bean (Figure 45).
Figure 45: Comparison of % bioavailable iron with the expected bioavailable iron in rice dishes containing Basmati rice and legumes.

Values are average of three replicates ± SD.

Figure 46: Comparison of % bioavailable zinc with the expected bioavailable zinc in rice dishes containing Basmati rice and legumes.

Values are average of three replicates ± SD.
6.3 CONCLUSION

Herbs used in Iranian rice dishes include dill, parsley and coriander and have a high iron content (4-5 mg/100g). These herbs were added to Basmati rice and all increased the bioavailability of iron in Basmati rice. The availability of iron in foods as consumed depends upon numerous factors, most of which are not fully understood. One factor often overlooked is the interaction of the iron with food during the digestion. As those herbs reported to be used in Iranian rice dishes contain vitamin C and several studies report that ascorbic acid will alter the bioavailable iron from cereal (Fairweather-Tait et al., 1995; Hunt et al., 1990; Hallberg et al., 1986). This increase in bioavailability may be due to a specific effect of the ascorbic acid in these herbs. Among herbs coriander was the most effective in increasing bioavailable iron (from 12.7% to 16.7%) while parsley was the most effective for increasing bioavailable zinc in rice dishes.

The bioavailable iron in the rice dishes containing Basmati rice and vegetables also increased with carrot and cauliflower being the most effective. This increase in the bioavailable iron may be due to the beta-carotene content of carrot and ascorbic acid in cauliflower. Garcia-Casal et al., (1998) reported in human study that vitamin A and beta-carotene can improve nonhaem iron absorption from rice.

Adding legumes including mung bean, lentil, broad bean and green bean increased bioavailable iron in Basmati rice. Lentil, broad bean and green bean were the most effective legumes to increase bioavailable iron while mung bean and lentil were the most effective to increase the bioavailable zinc. Among legumes
traditionally used in Iranian rice dishes, lentil was the most effective for increasing both bioavailable iron and zinc. Recent research has been conducted on the bioavailability of iron when consumed with various foods. In almost all cases the bioavailable iron between the different iron sources are changed, both increased and decreased. These changes are catalysed by the presence of certain foods or food ingredients, such as the enhancement of iron absorption, by the presence of meat or by the presence of vitamin C, or by certain vegetables rich in vitamin C and/or other vitamins to enhance the bioavailability or the inhibition of iron uptake by the presence of food rich in oxalate or phytate. The results from this study confirm that adding certain herbs, vegetables or legumes to Basmati rice could significantly increase the bioavailable iron as well as bioavailable zinc of the dish. Therefore if it can be established that improved availability of the iron depends upon the chemical environment of the iron in the food itself, a food may thus be designed or selected in which the iron is maximally available.
CHAPTER 7

7 CONCLUSION

7.1 GENERAL DISCUSSION AND CONCLUSION

Trace elements play a significant role in the aetiology of human disease. Consequently more extensive investigations of the relationship between the diet and mineral deficiencies needs to be promoted particularly in areas where dietary habits may lead to a significant risk to human health due to the development of trace element deficiency, imbalance, or excess. A successful diet must be compatible with human biological needs (Bryant et al., 1985). It must provide enough essential nutrients as well as energy to live and reproduce.

Members of cultural groups around the world share ideas about the manner in which food should be prepared. Nutritional changes during cooking or processing are numerous, taking place before, during and after food preparation. Bryant, et al. (1985) stated that an awareness of food processing effects on nutrient content is important because cooking and/or processing techniques can often make or break the overall quality of a group’s diet. For example maize is deficient in the B vitamin, niacin and the amino acid, tryptophan. There is a high prevalence of pellagra in many parts of the
world where corn or maize is used as a staple diet (Bryant et al., 1985). In some
cultures which use maize as a staple such as in North and South American, pellagra is
not evident due to the indigenous method used to process the maize. Katz (1983)
stated that the maize soaked in lime-water, wood ash or any other alkali solution
before grinding used traditionally by North and South American makes more niacin
and tryptophan bioavailable and thus prevents pellagra. The nutritional benefits of
traditional food processing techniques used in other cultures have not always been well
understood. For example Bryant et al. (1985) stated that Mexican American children
were encouraged by some nutrition educators to use white bread instead of maize.
Bryant et al. (1985) noted that not all processing techniques can enhance foods
nutritional quality. The traditional method of cooking rice in Iran has been examined
to understand the amount of bioavailable iron and zinc as well as phytate in the rice
meal prepared by traditional methods.

The available iron and zinc as measured by an in vitro method was shown to
increase when phytate was reduced. Beard et al. (1988) with human studies reported
that phytate decreased the absorption of iron from diets. Sandberg (1991) also
reported that phytate is one of the major inhibiting factors for zinc and iron absorption
according to in vitro measurement and human studies. Sharma and Khetarpaul, (1997)
found it imperative to reduce the level of anti-nutrients to improve the bioavailability of
the minerals in the human system.

Soaking in salted water reduced phytate level up to 53%; it also reduced the
total iron and zinc by 56% and 38%, respectively. For the remaining iron and zinc, the
bioavailability increased up to 41 and 40%; respectively. Oatway (2000) stated that
The total amount of any substances is not as important as the amount that is readily available for absorption.

Appropriate cooking of Basmati rice would increase its minerals bioavailability without any side effects and is economically efficient. This information could be incorporated into health and nutrition education messages aimed at increasing the utilisation of the rice in general.

In the present study, the bioavailable iron and zinc in the traditional Iranian rice meals were compared. The type of rice meal chosen was composed of Basmati rice and added herbs, vegetables or legumes. The addition of herbs such as coriander, due to the large ascorbic content, was also effective in improving iron absorption. Among vegetables, cauliflower was most effective for increasing the iron absorption of the rice. Again, cauliflower is a good source of ascorbic acid (Holland et al., 1991). Hallberg and Rossander (1984) stated that ascorbic acid is one of the main dietary enhancers of non-haem iron absorption and addition of foods rich in ascorbic acid may be one of the most effective ways of improving iron nutrition.

An increase in the bioavailable iron and zinc of a rice meal by the appropriate methods of cooking is one of the main techniques. An alternative approach is also to increase the bioavailability of iron in the rice meal by modifying the meal composition. An example would be the addition of foods that promote the non-haem iron absorption such as herbs or vegetables rich in ascorbic acid, carotene and/or riboflavin. Tuntawiroon et al. (1990) stated that the inhibitory effect of the phytate in the rice could be overcome by addition of rich ascorbic acid green leafy vegetables.
Cooking via the boiling method after soaking of rice in salted water removed 55% of the phytate but due to reduction of the huge amount of iron (56%) and zinc (38%), the overall bioavailable iron and zinc only increased to 23 and 16% of the original total, respectively. Mills et al. (1988) also reported that the total amount of zinc and iron intake as well as phytate are important for the availability of the zinc and iron. The results obtained from this research suggest that the traditional method of soaking and cooking of Basmati rice resulted in marked improvement of iron and zinc bioavailability.

Although fortification or enrichment of food with minerals and other nutrients is a well accepted practice that has been employed by food processors for more than half a century, Reilly (1996) reported that the acceptability of food fortification or enrichment of food has been questioned by regulatory authorities and some consumers. Despite fortifications and the progress that has been made in understanding the factors controlling iron absorption, iron and zinc deficiency continue to be a major nutritional problem in most developing countries.

Lawrence (1999) stated that taking a soybean gene for the iron storage protein, ferritin, and transferring this gene into the rice plants is a possible way of producing a grain of rice which will contain three times more iron than usual. One concern is with the other minerals; will this iron rich rice have any other effect on the bioavailability of its other minerals. Fisher et al. (1984) proposed that the consequence of excessive mineral intake has to be considered because it has been demonstrated that a daily intake of excessive zinc may alter the body’s copper status, with subsequent possible
risk of heart disease. Also, increasing iron intakes by super-enrichment of cereal products would inevitably, precipitate iron overload in some members of the population (Crosby, 1977). Walton (1999) stated that a well planned diet provides adequate iron. A good diet will safely help decrease the risk of inadequate iron and at the same time cause the least potential damage to those at risk for iron excess.

In spite of high amounts of iron and zinc in rice (Hallberg, 1974) bioavailability of these minerals is only about 12% of the total due to the high amounts of phytate. Phytate content of rice not only affects iron absorption from the rice, but also the iron absorption from other food in the same meal. Approximately 70% of the phytate comes from cereal (Davies, 1981) and by reducing the phytate content of cereal not only the absorption of iron and zinc from cereal may increase but also the absorption of iron from other food in the same meal, without any side effect or overload of minerals.

These research results confirm that soaking in salted water is an effective way to remove a significant proportion of phytate-phosphorus thereby improving the nutritive value of Basmati rice with regard to its positive effect on the bioavailability of iron and zinc.

Traditional cooking in this Iranian example enhanced iron and zinc absorption in a diet which was relatively low in these two minerals. There should be a high correlation between the evaluation of the composition of the traditional meal used by different ethnic groups and the iron status of that society. Investigation of the
bioavailable iron and zinc in traditional ethnic meals can provide valuable information as well as an important communication link between nutritionists and social scientists.

### 7.2 FURTHER RESEARCH

Further research will be necessary to evaluate the content of potent inhibitors and enhancers of iron absorption and their respective effects on iron solubility in typical traditional diets of Iranians. Evaluating the potential iron bioavailability in staple diets of Iranian is an important pre requisite for effective dietary modification which could improve the local diet and the nutritional status of that population. Decreasing inhibitors and increasing the intake of food containing ascorbic acid or other iron and zinc enhancers must be encouraged in this population since this may provide a potential solution to iron and zinc deficiency.
# GLOSSARY OF SYMBOLS AND ABBREVIATIONS

<table>
<thead>
<tr>
<th><strong>A</strong></th>
<th>Singlet oxygen acceptor</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Availability/bioavailability</strong></td>
<td>The term availability is considered synonymous with bioavailability which has been defined as the ratio between the quantity of iron in a sample as determined by biological assay to the quantity determined by chemical analysis.</td>
</tr>
<tr>
<td><strong>Basmati rice</strong></td>
<td>A variety of rice.</td>
</tr>
<tr>
<td><strong>Brown rice</strong></td>
<td>Rice grain which only the hull is removed and leaving is what we call brown rice. In brown rice the intact kernel covered with the bran layers.</td>
</tr>
<tr>
<td><strong>Cereals/Grains</strong></td>
<td>From ‘ceres’, the Roman goddess of agriculture and including all plants in the grass family such as rice, wheat, barley, oat, rye, corn and maize.</td>
</tr>
<tr>
<td><strong>Chelo</strong></td>
<td>Boiled and steamed white rice.</td>
</tr>
<tr>
<td><strong>e</strong></td>
<td>Molar extinction</td>
</tr>
<tr>
<td><strong>g</strong></td>
<td>Gram</td>
</tr>
<tr>
<td><strong>Gillian</strong></td>
<td>A State in the North of Iran.</td>
</tr>
<tr>
<td><strong>h</strong></td>
<td>Hour</td>
</tr>
<tr>
<td><strong>I-</strong></td>
<td>Incident intensity</td>
</tr>
<tr>
<td><strong>o</strong></td>
<td>Degree</td>
</tr>
<tr>
<td><strong>Kg</strong></td>
<td>Kilogram</td>
</tr>
<tr>
<td><strong>L</strong></td>
<td>Litre</td>
</tr>
<tr>
<td><strong>ln</strong></td>
<td>Refractive index</td>
</tr>
<tr>
<td><strong>NSW</strong></td>
<td>New South Wales</td>
</tr>
<tr>
<td><strong>p-</strong></td>
<td>Incident intensity</td>
</tr>
<tr>
<td><strong>p</strong></td>
<td>Probability</td>
</tr>
<tr>
<td>Term</td>
<td>Definition</td>
</tr>
<tr>
<td>------</td>
<td>------------</td>
</tr>
<tr>
<td>Phytate</td>
<td>Phytate is the calcium salt of phytic acid (Myo-inositol hexakisphosphate, IP6). A free acid of phytic acid called phytate or phytin exist according to the physiological pH and the metal salts present. All these forms are used interchangeably in most of the literature.</td>
</tr>
<tr>
<td>Polow</td>
<td>White rice mixed with different ingredient such as herbs, vegetables or legumes.</td>
</tr>
<tr>
<td>SD</td>
<td>Standard deviation.</td>
</tr>
<tr>
<td>SE</td>
<td>Standard error.</td>
</tr>
<tr>
<td>Seljuq</td>
<td>One of the ancient kingdom in Persia. A family of Turkish mercenary soldiers that rose to prominence and conquered much of Asia minor in the 11 and 12 century. They were converted to the Muslim faith and became established as sultans in the area of present day Syria and eastern Turkey.</td>
</tr>
<tr>
<td>Qajar</td>
<td>One of the ancient kingdom in Persia. Qajar a Turcoman tribe which furnished Persia with a dynasty which ruled from 795 to 1924.</td>
</tr>
<tr>
<td>W/V</td>
<td>Weight/Volume</td>
</tr>
</tbody>
</table>
Appendices
CONSENT FORM

UNIVERSITY OF WESTERN SYDNEY, HAWKESBURY

This project is designed to provide information for future program planning in nutrition and health. Your personal data is important for planning nutrition advice programs to meet the needs of the people. This study will be conducted through the use of 24 unit questionnaire. All information and data obtained will be treated confidentially. The inconvenience to respondents, including the time taken to conduct the interview, is estimated to be 10 minutes.

Volunteers Consent

I have been briefed on this research project and I give my consent. By signing this form I understand that information provided by me will be confidential and that I may withdraw at any time without penalty.

FULL NAME-----------------------------

SIGNATURE-----------------------------

DATE-----------------------------
Questionnaire

1- Male
   Female

2- Which of these age groups do you belong to?
   Under 25
   26 - 35
   36 - 45
   46 +

3- Where were you born?

4- How long have you been in Australia?

5- What is the highest level of education you have completed?
   None
   Primary school
   Secondary school
   University/College

6- What is your current employment status?
   Employed full time
   Employed part time
   Unemployed
   Student

7- How often do you cook rice?
   Every day
   2-3 times a week
   once a week
   Others
8- Do you wash your rice before cooking?

   Yes -----  
   No ----- 

If yes how many wash?

9- Do you soak your rice before cooking?

   Yes ----- 
   No ----- 

If yes for how long?

   Under 2 hours -----  
   2 hours ----- 
   4 hours ----- 
   8 hours ----- 
   12 hours ----- 
   24 hours ----- 
   Others ----- 

10- Do you add salt to your soaking water?

   Yes ----- 
   No ----- 

11- How do you cook your rice?

   Absorption method ----- 
   Boiling method ----- 
   Others ----- 

250
12- How do you serve your rice dish?

<table>
<thead>
<tr>
<th></th>
<th>Daily</th>
<th>2-3/week</th>
<th>Weekly</th>
<th>Rarely</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plain rice</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Plain rice with meat</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Plain rice with stew</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Plain rice with white meat</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Plain rice with red meat</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td>Mixed rice (rice and vegetable)</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
<td>-----</td>
</tr>
</tbody>
</table>

Please specify.

| Mixed rice (rice and beans) | ----- | ----- | ----- | ----- | ----- |
| Please specify. |

13- In the last years, has your consumption of rice increased, decreased or remained unchanged?

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Increased</td>
<td>-----</td>
</tr>
<tr>
<td>Decreased</td>
<td>-----</td>
</tr>
<tr>
<td>Remained unchanged</td>
<td>-----</td>
</tr>
</tbody>
</table>

14- For which of the following reasons do you usually consume rice?

<table>
<thead>
<tr>
<th>Reason</th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td>You like it</td>
<td>-----</td>
<td>----</td>
</tr>
<tr>
<td>Nutritional value</td>
<td>-----</td>
<td>----</td>
</tr>
<tr>
<td>Tradition</td>
<td>-----</td>
<td>----</td>
</tr>
<tr>
<td>Availability</td>
<td>-----</td>
<td>----</td>
</tr>
<tr>
<td>Convenience</td>
<td>-----</td>
<td>----</td>
</tr>
<tr>
<td>Cost</td>
<td>-----</td>
<td>----</td>
</tr>
<tr>
<td>Others</td>
<td>-----</td>
<td>----</td>
</tr>
</tbody>
</table>

15- Do brand names influence your choice of rice? If yes, what brand do you usually buy?

<p>| |</p>
<table>
<thead>
<tr>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
</tr>
<tr>
<td>No</td>
</tr>
</tbody>
</table>
16- Where do you usually purchase your rice?

<table>
<thead>
<tr>
<th></th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td>Supermarket</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Iranian shops</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Others (please specify)</td>
<td>------</td>
<td>------</td>
</tr>
</tbody>
</table>

17- What kind of pan do you usually use to cook your rice?

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th>----</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stainless steal</td>
<td>----</td>
</tr>
<tr>
<td>Non stick pan</td>
<td>----</td>
</tr>
<tr>
<td>Aluminium pan</td>
<td>----</td>
</tr>
<tr>
<td>Others</td>
<td>----</td>
</tr>
</tbody>
</table>

18- How often do you eat the following?

<table>
<thead>
<tr>
<th></th>
<th>Daily</th>
<th>2-3/week</th>
<th>Weekly</th>
<th>Rarely</th>
<th>Never</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fruits</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fresh</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Dried</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Canned</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Vegetable</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fresh</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Dried</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Meats</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Red</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>White</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Fish</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Rice</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Brown</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Drinks</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tea</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Coffee</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
<tr>
<td>Water</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
<td>----</td>
</tr>
</tbody>
</table>
19- Which food do you consider as being a good source of iron or zinc?

<table>
<thead>
<tr>
<th></th>
<th>Iron</th>
<th></th>
<th>Zinc</th>
<th></th>
<th>Do not know</th>
</tr>
</thead>
<tbody>
<tr>
<td>Red meat</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>White meat</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Vegetables</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Fruits</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Cereals</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Pulse</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
</tbody>
</table>

20- Which groups do you think are at highest risk for iron or zinc deficiency?

<table>
<thead>
<tr>
<th></th>
<th>Iron</th>
<th></th>
<th>Zinc</th>
<th></th>
<th>Do not know</th>
</tr>
</thead>
<tbody>
<tr>
<td>Women</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Pregnant women</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Men</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Teenagers</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Children</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Vegetarian</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Meat eater</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
</tbody>
</table>

21- Do you take any vitamin or mineral supplements? (if yes please specify).

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>----</td>
</tr>
<tr>
<td>No</td>
<td>----</td>
</tr>
</tbody>
</table>

22- Which of the following items, do you think may increase iron or zinc absorption of the meal? please tick as many as you wish

<table>
<thead>
<tr>
<th></th>
<th>Increase</th>
<th>Reduce</th>
<th>Dont know</th>
</tr>
</thead>
<tbody>
<tr>
<td>Red meats</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Beans</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Nuts</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Cereals</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Fruits</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Vegetables</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Orange juice</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Tea &amp; coffee</td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
</tbody>
</table>
23- Do you feel:

- Energetic
- Tired
- Lethargic

24- Do you have:

- Reduce ability to exercise
- Poor stamina
- Frequent infection
- Anaemia

Thank you for your time
Phytic acid determination

Procedure

1. Weigh 2g of sample into a 100ml conical flask and add 40 ml of 0.38M HCl/10% Na₂SO₄.
2. Add a magnetic flea to flask and extract for 3 hours on a magnetic stirrer.
3. Filter through No. 1 Whatman filter paper.
4. Pipett 5.0ml of filterate into centrifuge tubes and add 5.0ml water and 6.0ml of 0.4% FeCl₃ 6H₂O in 0.075M HCl.
5. Heat tubes in boiling water bath for 15 minutes.
6. Centrifuge tubes on high for 15 minutes, then decant and discard supernatants.
7. Disperse each pellet thoroughly with 5ml 4% Na₂SO₄ in 0.075M HCl.
8. Centrifuge for 15 minutes on high, then decant and discard supernatants.
9. Dissolve the pellet in 3mL Conc. H₂SO₄, then add 3mL water. Transfer to glass test tubes contain a few antibumpers granules.
10. Place in a sand bath and hold at boiling point for 90 minutes (180° C).
11. Add 15 mL of water to test tubes.
12. Heat in a boiling water bath for 15 minutes.
13. Qualitatively transfer to 50 mL volumetric flask and dilute to volume with DW.
14. Determine phosphorus by taking a suitable aliquot measuring spectrophotometrically the colour reaction of phosphomolybdate with ANSA reagent.

Principle

After extraction of the sample, phytic acid (inositol hexaphosphate) is precipitated as ferric phytate. The phosphorus content of the hydrolysed precipitate is measured and phytic acid calculated.
**Calculation**

\[
\text{Mg/100g Phosphorus} = \text{sample reading} \times \text{Std factor} \times \frac{40}{5} \times \frac{50}{2} \text{ aliquot taken}
\]

\[
\text{Mg/100g Phytic acid} = \frac{\text{mg/100g phosphorus} \times \text{molecular wt Phytic acid}}{\text{Molecular wt P} \times 6}
\]

ie. \[
\text{Mg/100g Phytic acid} = \frac{\text{mg/100g P} \times 660}{186}
\]

**Reference**

Caldwell, University of Sydney, 1991 as provided by the Bread Research Institute (BRI). Riverside Corporate Park, West Entrance Delhi Road, North Ryde, NSW 2113.
Total iron

Spectrophotometric method

Determination:
- Ash 10g rice flour in porcelain dish
- Moisten ash with 0.5-1.0ml Mg(No3)2 soln.
- Dry and carefully ignite in furnace
- Cool, add 5ml HCL, letting acid rinse upper portion of dish and evaporates to dryness on steam bath
- Dissolve residue by adding 2.0ml HCL and heat 5 min on steam bath with watch glass on dish
- Rinse watch glass with H2O, filter into 100ml vol. Flask, cool and dilute to volume
- Pipet 10ml aliquot into 25ml vol. Flask and add 1 ml H2NOH.HCL soln. in few min add 5 ml buffer soln. and 2ml dipyridyl soln. And dilute to volume
- Determine total iron using Atomic absorption spectrophotometer

Ref: AOAC (Official Methods of Analysis (1990)

Availability of iron and zinc

Two grams of powdered rice was incubated by pepsin-HCL solution (0.5% pepsin in 0.1 N HCL). The mixture was adjusted to pH 1.3 and incubated in a 100ml conical flask at 37°C in a metabolic shaker water-bath for 2h and subsequently the pH was raised to 7.5 before incubating for another 2h. At the end of this incubation period the contents of the flask were centrifuged at 5000rpm for 30min, and the supernatant containing the free form of these elements was collected. An aliquot in suitable dilution was placed in an atomic absorption spectrophotometer to determine ionisable Fe and Zn.

REFERENCES


Darby, W. J. (1972). The case for the proposed increase in iron enrichment of flour and wheat products, Nutrition Review. 30 (4): 98.


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Suharno, D. West, C. E. Muhilal, Karyadi, D. and Hautvast, J. A. J. (1993). Not only supplementation with iron but also with vitamin A is necessary to combat nutritional anemia in pregnant women in West Java, Indonesia. Lancet. 342: 1325-1328.


