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Phytate and minerals in Indian baby foods

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PHYTATE AND MINERALS IN INDIAN BABY FOODS

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ABSTRACT

PHYTATE AND MINERALS IN INDIAN BABY FOODS

By Kavita Sharma

The objective of this study was to analyze five commercially available Indian baby foods for phytate, calcium, iron, and zinc. Molar ratios of phytate/calcium, phytate/iron, phytate/zinc were calculated to determined the bioavailability of these essential minerals.

The phytate content (mg/100g) ranged from 1.22 (Dexolac) - 1.97 (Farex). Iron content (mg/100gm) ranged from 8.0 (Cerelac Wheat) - 10.0 (Farex). Zinc content (mg/100gm) ranged from 2.8 (Nestrum) - 8.5 (Cerelac Wheat) and calcium content (mg/100gm) ranged from 97.5 (Nestrum) - 333.0 (Dexolac). Molar ratios of phytate/iron ranged from 0.006 (Nestrum) - 0.018 (Cerelac Wheat). Molar ratios of phytate/zinc ranged from 0.019 (Cerelac Wheat) - 0.045 (Nestrum). Molar ratios of phytate/calcium ranged from 0.0002 (Dexolac) - 0.0008 (Nestrum). Molar ratios of phytate/zinc and phytate/calcium were well below the published value associated with adequate zinc and calcium bioavailability of 10 and 0.2 respectively. The results showed that, the phytate content of the baby foods was unlikely to inhibit bioavailability of zinc and calcium. As phytate concentration was low in all the baby foods analyzed, it was concluded that iron was also bioavailable.

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PREFACE

The following thesis is written in publication style. The second chapter is written in journal format and will be submitted to The International Journal of Food Science and Nutrition. Chapters 1 and 3 are written according to the guidelines outlined in the Publication Manual of the American Psychological Association, 4^{th} edition, 1995.

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CHAPTER 1

INTRODUCTION AND REVIEW OF LITERATURE

Introduction

The best food for infants is human milk. In the first three or four months of an infant's life, a period of rapid growth, most infants thrive well on their mother's milk alone. However, by five to six months of age, the supply of energy and some nutrients from human milk are no longer adequate to meet the growth requirements. The infant needs other foods besides milk which supply energy, protein, minerals, and other nutrients. It has been reported that early weaning is a common practice in India and many developed countries where socio-economic levels are high (Dodd & Datta, 1988). In a study conducted by Gibson, Ferguson, and Lehrfield (1998), the complementary foods used in developing countries were reported to be mainly from cereals or starchy roots and tubers. They are prepared as thin gruels. The energy and nutrient contents of these gruels are low. It is further exacerbated if the infants receive very few feedings per day.

Malnutrition is widespread in India and many developing countries, and the condition is particularly serious in children below 3 years of age. It is estimated that between 30% to 50% of the children in this age group are malnourished (Ashturkar, Pande, & Reddy, 1992). Malnutrition in many regions may not be due to energy and protein deficiency alone. It may be a cumulative effect of many factors such as large families, poverty, ignorance, deficient diets, infection, or poor absorption of minerals (Sehgal, Kapoor, & Taneja, 1989).

In India, legumes and whole grain cereals are the main and most economic source of nutrients such as protein, vitamins, carbohydrates, and minerals like calcium, magnesium, iron, and zinc in an infant's diet. A major portion of the phosphorous content of these plant-based foods is phytate phosphorus. Phytate is a complex salt of calcium and magnesium with myo-inositol and is the principal storage form of phosphorus in many kinds of seeds (Pawar & Ingle, 1987). Phytate ion complexes with metallic ions like zinc, calcium, magnesium, and iron that form insoluble compounds and interfere with mineral utilization. Formation of insoluble compounds results in a decrease in mineral bioavailability, particularly in infants when periods of growth are rapid.

Iron deficiency is a worldwide problem affecting a large number of people in western communities as well as in underdeveloped countries. It is more prevalent in infants, and is usually the result of low bioavailability of dietary iron (Fox, Eagles, & Fairweather-Trait, 1998). Infants and children have higher needs for the zinc because they are growing rapidly and synthesizing many zinc containing proteins (Whitney & Rolfes, 1993a). Calcium malnutrition in the form of rickets affects a wide number of infants. Although minerals are needed in small amounts, they play important roles in an infant's body.

Objective

The objective of this present study was to determine the phytate, calcium, iron, and zinc contents of five commercially available Indian baby foods. Molar ratios of phytate/calcium, phytate/iron, and phytate/zinc were determined to estimate bioavailability of calcium, iron, and zinc in the five baby foods.

Significance of the Study

In India plant foods like whole grain cereals, legumes, and fruits form the bulk of the infant's diet due to religious beliefs or for socio-economic reasons. These plant foods are the major and economic source of minerals and other nutrients, and prepare infants for a more varied diet. At present, commercially prepared baby foods are widely used in India for an infant's diet. The growing tendency to use commercial baby foods necessitates increased research on their nutritional quality.

Phytate is a naturally occurring substance in the plant kingdom that is found in highest amounts in cereal grains and legumes. It binds minerals in the gastrointestinal tract by forming insoluble complexes. Formation of the phytate-mineral insoluble complex results in a decrease in mineral bioavailability, particularly in infants when periods of growth are rapid. The increased use of plant-based foods in an infant's diet could be a problem related to bioavailability of minerals due to the presence of phytate.

The information on the effect of phytate on bioavailability of iron, calcium, and zinc in Indian baby foods is limited. The effect of phytate on mineral bioavailability is probably useful in estimating the molar ratios of phytate/iron, phytate/calcium, and phytate/zinc. Therefore molar ratios are estimated in the present study to determine the bioavailability of minerals in Indian baby foods.

Results of the present study can be useful in determining the bioavailability of iron, calcium, and zinc from commercially prepared Indian baby foods. Based on knowledge of the bioavailability of minerals of these baby foods, new improved baby foods can be developed for infants with high bioavailability of minerals.

Review of literature

Naturally occurring important phosphorus compounds in plant foods (especially in cereal grains, legumes, and beans) are phytic acid, and its salts, phytate. (Erdman & Forbes, 1977).

Chemistry of Phytate

Numerous polyphosphorylated inositols can be found in nature, and depending on the complex formed, a wide variety of compounds can exist, such as phytin, phytate, phytates and phytic acid. Phytic acid is commonly called myo-inositol hexaphosphoric acid or significantly 1,2,3,4,5,6 – hexakis (dihydrogen phosphate) myo-inositol. The term phytin implies a calcium-magnesium salt of phytic acid, whereas phytate refers to the mono to dodeca anion of phytic acid (Maga, 1982). Commercially available phytic acid or phytate often contains lower phosphate derivatives than hexphosphate (Cosgrove, 1963), which some investigators include in the term phytates (Maga, 1982). There are nine sterisometric inositoles possible, namely cis-inositol, epi- inositol, allo- inositol, neo inositol, myo- inositol, muco- inositol, chiro- inositol, and scyllo- inositol. Out of all of these, only the myo form has been isolated from plants, while neo, chiro and scyllo-inositol hexaphosphates have been identified in soils (Cosgrove, 1966).

Although the conformation of the phytic acid molecule is still in question (Maga, 1982), the most acceptable structure of phytic acid, the hexaphosphate of myo- inositol which was suggested by Anderson (Anderson, 1914), is shown in Figure 1. The results of potentiometric titration of crystalline sodium phytate supports the Anderson Structure for phytic acid (Erdman, 1979).

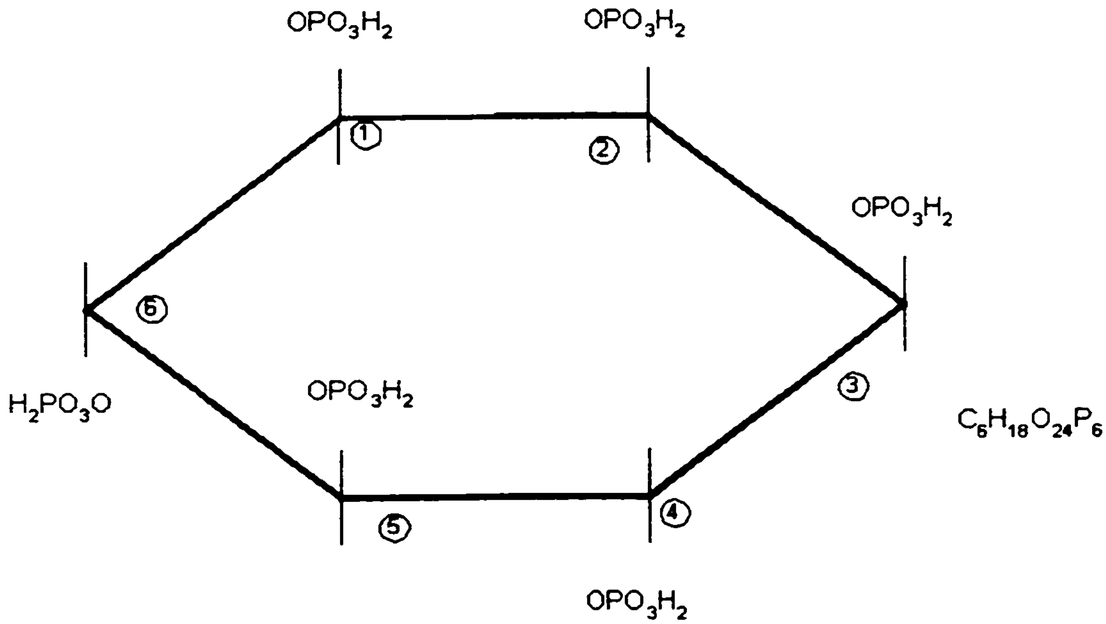

Figure 1: Anderson Structure for Phytic Acid.
Note: From "Oilseed phytates: Nutritional implications" by J. W. Erdman, Jr., 1979, Journal of American Oil Chemists' Society 56, p. 736.

Antinutritional Aspects of Phytate

Phytate (inositol hexaphosphate) represents a complex phosphorus compound that can significantly influence the functional and nutritional properties of the food (Maga, 1982). Phytate has a strong metal chelation ability, which is pH dependent. Inositol hexaphosphate and some of its degradation products, can interfere with mineral absorption by forming insoluble mineral inositol phosphate complexes. Most of the essential minerals and trace elements are absorbed in the duodenal or jejunal part of the small intestine. The site and degree of phytate degradation can affect the nutritional value of a high phytate diet.

Phytate inhibits mineral absorption because the negative charges of the phosphate groups form insoluble salts upon interaction with divalent cations. Due to its ability to chelate with metal ions, it often impairs the dietary bioavailability of metals such as iron, calcium, and zinc in foods for human and animal nutrition (Erdman & Forbes, 1977).

In most legume seeds, phytate phosphorus accounts for approximately 80% of the total phosphorus and is present primarily as a metal phytate or complexed with proteins. Phytate is a potential source of phosphorus, which can be liberated during the germinating stage of the seeds. Phosphorous is readily absorbed from the diet in the form of free inorganic phosphate after digestive hydrolysis. Only the phosphorous that is part of dietary phytic acid or phytates is not well hydrolyzed and is therefore unavailable. Phytic acid forms insoluble calcium, magnesium, and zinc salts preventing absorption (Linder, 1991). Furthermore some of the isomers of inositol phosphates formed during degradation have important physiological functions in humans and animals. According to

Harland and Morris (1995), the mineral binding strength becomes progressively lower as the solubility increases when phosphate groups are removed from the inositol hexaphosphate.

Dephosphorylated products of phytate, which result from endogenous or exogenous phytase, do not exhibit the inhibitory effect of phytate on mineral absorption. These compounds can be separated by anion exchange chromatography. The 1,2,3-triphosphate grouping increases iron solubility and 1,2,3,6-tetraphosphate increases calcium absorption. Thus, intermediates of phytate hydrolysis may enhance mineral absorption (Weaver, 1998).

Studies show that phytic acid and its derivatives decrease the bioavailability of many nutritionally essential minerals such as calcium, magnesium, iron, and zinc (Erdman & Thompson, 1982; Maga, 1982; Serraino & Thompson, 1984). These complex formations between phytate and minerals have been reported to interfere with the enzymatic degradation of proteins (O' Dell & deBoland, 1976; Singh & Krikorian, 1982).

The relative binding affinity by phytate for minerals was studied by Vohra, Gray & Kratzer (1965). Using the titrimetric method, they found that the capacity of sodium phytate to form a complex with a metal is pH dependent. The binding capacity of phytate to the metals at pH 7.4 appeared to be in the following decreasing order, copper, zinc, nickel, cobalt, manganese, iron, and calcium.

Chelation

The term chelate is derived from chele, the Greek word for claw. A chelating agent is a molecule that is capable of seizing and holding a metal ion in a claw-like grip.

The clutching structure forms a ring in which the ion is held by a ligand (Clydesdale, 1988). A chelate is a complex resulting from the combination of a metal ion and a multidentate ligand such that the ligand forms two or more bonds with the metal. The resulting structure is a ring that includes a metal ion (Fennema,1996).

The metal atom (cation) in a chelation reaction is referred to as the central atom. The molecules (anions) that have an unshared electron pair(s), which can be donated to the central atom to form a coordinate covalent bond are known as coordinating groups or ligands. The number of bonds formed by the central atom is called its coordination number. Some minerals have only one coordination number and form complex ions.

Phytic acid has a tremendous chelating potential due to the presence of twelve ionisable hydrogen atoms under physiological pH conditions. Phytic acid is extensively ionized and is capable of interacting strongly with metal ions and proteins (Nolan, Duffin & McWeeny, 1987). Phytate being a strong ligand, can form stable complexes with various metal cations. Various cations can strongly chelate between two phosphate groups or within a phosphate group. In a mixed chelate of phytic acid, the relative binding strengths of various metal ions to phytic acid differ greatly (Erdman, 1979).

Depending on the complex formed between phytate and metals, a variety of structures are possible. The phosphate group may chelate to give 4-membered ring complexes as shown in Figure 2A. The structure where two or more phosphate groups from the same or from different phytate ions may complex to one metal cation is shown on Figure 2B. The polymeric structure is shown in Figure 2C. This is the complex

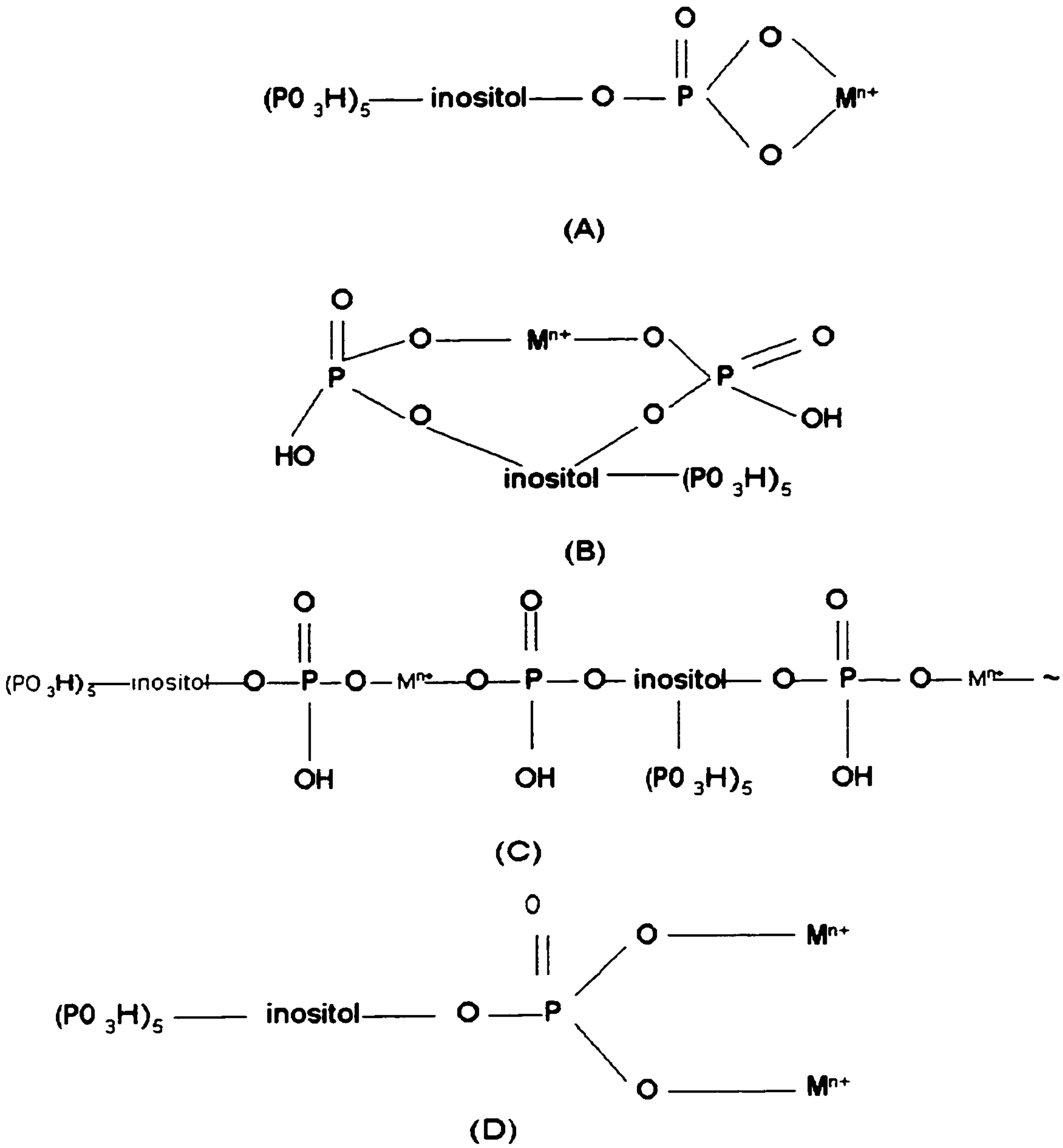

Figure 2 . Structure of phytic acid Chelate
Note :From "Effects of Phyate on Mineral Bioavailability . In Vitro Studies on Mg2+, Ca 2+, Fe 3+, Cu 2+, and Zn 2+ (also Cd 2+) Solubilities in the presence of Phytate" by K. B. Nolan, P. A. Duffin, & D. J. McWeeny, 1987, Journal of Science and Food Agriculture , 40, p. 81

between two phytate ions and a metal. A phosphate group from phytate may also serve to bridge two metal ions as shown in Figure 2D.

Occurrence of Phytate

The organic compound phytate is naturally present in a wide variety of foods like legumes, cereal grains, nuts, seeds, some fruits and vegetables and possibly in all cells of the plants (Harland & Morris, 1995). Phytates have been found in cereal grains and legumes up to a level of approximately 5% by weight (deBoland, Garner & O'Dell, 1975). In most seeds, phytic acid is associated with specific components and can be extracted with those components. For example, the endosperm of wheat and rice kernels are almost devoid of phytate as it is concentrated in the germ and aleurone layers of cells of the kernels and in the bran or hull. The phytic acid concentrations in morphological components of wheat and rice are shown in Table 1.

Asada, Tanaka & Kasai (1978) reported that during the early stage of rice ripening a major portion of the myo-inositol was in the free state, but at the end of the ripening period most of the myo-inositol was in the phosphate ester form which represented approximately 80% of the total phosphorus in the product. The amount of phytic acid formed in rice grains was directly affected by the amount of phosphate available in the plant, and they postulated that excess phosphate could be stored in the form of phytic acid or phytate. Thus, excessive phosphate fertilization could result in high phytate levels.

Kennedy and Schelstraete (1975) reported that phytic acid was primarily found in the outer layers of rice grain. Specifically, 2% of the outside kernel was found to contain

Table 1: Phytic Acid Concentration in Morphological Components of Cereals

Sample		Phytic acid %
Wheat	Soft	1.13
	Germ	3.90
	Endosperm	< 0.01
	Aleurone	4.11
	Hull	None Detected
Rice	Brown	0.89
	Germ	3.48
	Endosperm	0.01
	Pericarp	3.37

Note: From "Oilseed Phytates: Nutritional Implications" by J. W. Erdman Jr., Journal of American Oil Chemists' Society, 1979, p.738.

23 times more phytic acid than the intact kernel, and the removal of the outer 13% of the kernel resulted in an endosperm that contained no detectable phytic acid. Thus, if one is specifically concerned about phytic acid levels in rice, milling can be used to decrease its level.

In evaluation of 38 wheat varieties, Lolas, Palamidas & Markakis (1976) found a phytic acid range of 0.62%- 1.35% dry weight in wheat kernels whereas the bran portion had phytic acid levels ranging from 4.59 to 5.52 %, demonstrating that foods containing added wheat bran could have unexpectedly high levels.

Phytic acid levels in 18 varieties of barley were found to range from 0.97% to 1.08% dry weight (Lolas et al., 1976). Lolas and Markakis (1975) measured the phytic acid level of 50 cultivated varieties of Phaseolus Vulgaris (dry beans) and found a range of 0.54% - 1.58%. They also found a high degree of correlation between total phosphorous content and phytic acid level. In addition, a protein-phytate complex was isolated. It was noted that over 99% of the total phytic acid was in a water soluble form which could serve as a means of removing or lowering phytic acid levels in Phaseolus Vulgaris.

Other products that have been shown to contain phytates include crested wheat grass (Wilson & Harris, 1966) potatoes (Samotus & Schwimmer, 1962), and Bengal gram seed (Naik & Narayana, 1959; Verma & Lal, 1966; Reddy & Salkunkhe, 1981). Oberleas (1973) reported that lettuce, onions, mushrooms, celery, and spinach do not contain phytates, while green beans, carrots, and broccoli have only trace amounts. Sweet potatoes and artichokes are known to contain phytate in moderate amounts while nuts, cereals, and legumes contain high levels. Apricots, canned apricots, dried banana, citrus pectins, canned fruit cocktail, grapefruit, kale, canned peaches, canned peaches in light syrup, dried peaches, raw pineapple, canned prunes, and spinach contain no detectable phytate (Harland & Morris, 1995).

Biological Function of Phytate

In almost all seeds, phytates are considered the chief storage form of phosphate and inositol. (Cosgrove, 1966). During the ripening process of seeds the phosphorus is transported to the seeds from the leaves and roots. Most of the transported phosphorus is eventually found in phytic acid. In cereal grains 60%-80% of the total phosphorus is accumulated in phytic acid. Formation of phytic acid during maturation of seeds and tubers is thought to prevent accumulation of excessively high levels of inorganic phosphate (Cosgrove, 1966). It has been generally assumed that phytic acid is utilized as a source of phosphorus at germination. Chen and Pan (1977), reported that there is a dramatic increase in the phytate activity 5 days after germination in soybeans and in a variety of peas.

As phytic acid accumulates in various storage sites in seeds and tubers, other minerals apparently chelate to it forming the complex salt phytate. (Cosgrove, 1966).

Methods of Removal of Phytate

Phytic acid is at least partially responsible for reduced bioavailability of certain minerals from high phytate foods, and monogastric animals such as humans have little or no intestinal phytase activity. It would be advantageous to develop methods to either remove phytic acid from cereals and legumes or to reduce its mineral binding capacity.

Extensive studies have been designed to develop ways to eliminate phytate from foods by soaking, extracting the foods (phytate is soluble in an acid solution) or by enhancing fermentation, thus creating phytate hydrolysis products, which have weak mineral binding properties (Lonnerdal, Jayawickrama & Lien, 1999;Harland & Morris, 1995). Dephosphorylation of phytic acid (hydrolysis) in bran increases the nutritive value of cereals. Other processing methods like milling, cooking, germination, autoclaving, refining, soaking, and boiling are also known to reduce or remove considerable amounts of phytate in foods. However, due to the processing of foods, significant amounts of minerals and vitamins are lost (Salunkhe & Kadam, 1989b). Phytic acid in infant cereals can be degraded by adding exogenous phytase (Davidsson, Galan & Cherouvrier, 1997).

Fermentation of cereals and legumes appreciably reduces the phytate content due to the presence of endogenous phytase and that of added yeast and other useful microorganisms. During fermentation of soybeans, Sudarmadji & Markakis (1977) observed that the decrease of phytic acid was accompanied by an increase in inorganic phosphate. They concluded that the reduction in phytic acid obtained was due to the action of the enzyme phytase, which was produced by mold during fermentation. Fermentation of whole-wheat flour by yeast was shown by Reinhold, Parsa, Karimian, Hammick & Ismail-Beigi (1975) to destroy a high proportion of phytates. The effectiveness and rate of destruction of phytates in whole wheat meals were higher in flours of low extraction rate.

Phytic acid in mung beans is not completely destroyed by cooking. About 58 to 85% of phytic acid remains after autoclaving for 30 minutes at 120 °C. Cooking mung beans in water containing 3% NaCl at 100 °C for 3 hours resulted in a slight decrease in phytic acid (Tabekhia & Luh, 1979).

Toma and Tabekhia (1979), reported that the water quality used during the cooking of rice can significantly influence phytic acid levels. Cooking for 10 minutes in domestic tap water resulted in no significant loss of phytic acid, however, when distilled deionized water was utilized, phytic acid content was reduced by approximately two-thirds. The ability of phytic acid to form salt complexes was cited as the reason for this difference.

Minerals Bioavailability

Minerals are inorganic nutrients that remain as ash when plant or animal tissues are burned. Although they contribute small amounts of body weight, they play important roles in the regulation of body fluid, acid base balance, and metabolic processes (Pipes, 1977). Defects in mineral nutrition are capable of producing severe impairment of health. The three elements mainly involved in this study are calcium, iron, and zinc. The chemical presence of a mineral provides little assurance of its availability for absorption and utilization. Minerals from cereals, legumes, oilseeds, and other plant foods compared to minerals from animal sources are poorly absorbed by human beings. Certain components present in plant foods are mineral binders and are known to chelate minerals and reduce their absorption. Some of the mineral binders other than phytate are dietary fiber, polysaccharides, oxalates and polyphenol compounds. These may also play a major role in mineral bioavailability.

Rackis and Anderson (1977) reported that reduced availability of essential minerals in the intestinal tract due to phytate depends on several factors such as, ability of endogenous carriers in the intestinal mucosa to absorb essential minerals bound to phytate and other dietary substances, concentration of phytic acid in the food, concentration of minerals in the food, digestion or hydrolysis of phytate by the phytase enzyme in the intestine, phytate inhibition, and processing of products or methods of processing.

Iron

Iron is essential for all living organisms since iron containing proteins have a crucial role in both oxygen transport in the respiratory chain and in DNA synthesis (Hemminiki, 1995). Iron deficiency is the most prevalent nutrition deficiency among infants and young children in industrialized as well as developing countries (Ziegler & Fomon, 1996). The prevalence of iron deficiency anemia in South East Asia is among the highest in the world (FAO/WHO, 1992; Scrimshaw, 1994). One of the major risk groups for iron deficiency anemia is children between the ages of 6 months and 18 months when growth velocity is still high, and human milk does not provide sufficient iron. This deficiency develops only after iron stores are seriously depleted and hemoglobin production slows until hemoglobin concentrations are below normal (Herbert, 1987). During this time infants require additional iron from food with high iron bioavailability (Lam, Gross, Sastramidjojo & Schultink, 1997). This deficiency in infants has been linked to impairment of intellectual development, which may be irreversible (Fox et al., 1998). Other deleterious effects of iron deficiency include fatigue, impaired ability to maintain body temperature in a cold environment, and behavioral abnormalities.

Since iron from breast milk alone is not sufficient, adequate supplementary feeding is needed in terms of quantity and quality (Lam et al., 1997). In India and other developing countries two of the most important sources of iron in infant food are infant cereals and legumes which are fairly good sources of iron, but have poor iron bioavailability. As previously mentioned, cereals and legumes contain phytate that is known to decrease bioavailability by binding with non heme iron. Phytic acid has the ability to bind up to 6 metal ions. Therefore it reduces the absorption of dietary iron because of the formation of insoluble complex (Simpson, Morris & Cook, 1981). Infants consuming only plant-based foods are at a greater risk for iron deficiency because non heme iron from plant foods is more poorly absorbed than heme iron from animal sources.

Phytate content of infant foods can be decreased by either enzymatic treatment (phytase) or by processing such as fermentation which improves iron absorption (Hallberg, Rossander & Kanberg, 1987). Absorption of iron from legumes commonly used in weaning foods like mung beans was observed to be only ~ 1%-2% (Lynch, Beard & Dassenko, 1984). In the US, about 20%-25% of all formulas consumed are soy formulas. Iron absorption from soy formula and soy-containing infant foods is low (Hallberg & Rossander, 1982; Morck, Lynch, Skikne & Cook, 1981; Lynch, Dassenko, Morck, Beard & Cook, 1985). This is because soy and other legumes, like cereals, contain phytate, which reduces the bioavailability of iron (Lynch et al., 1984).

Cook, Reddy, Burri, Juillerrat & Hunell (1997) studied the influence of different cereal grains on the iron absorption from infant cereal foods and concluded that the type of cereal grain has little influence on iron bioavailability of infant cereal. On the other hand, modifications in the milling and processing methods for cereal grains that reduce their content of phytate acid are likely to improve iron availability significantly. Iron content of fruits used in infant foods is usually low, but they may help to improve iron bioavailability of the diet as a whole. Most citrus fruits contain ascorbic acid, citric acid, and other organic acids which increase iron absorption by enhancing iron solubility (Gillooly, Bothwell & Tonance, 1984). The ability of ascorbic acid to overcome the inhibitory effect of phytic acid on iron bioavailability depends on both the native content of phytic acid in food and the amount of ascorbic acid added.

Zinc

Zinc is distributed throughout all cells and tissues. The highest concentrations are in the male sex gland, bone, and hair. Some of the roles of zinc in the body are making genetic material and proteins, taste perception, and involvement in wound healing (Whitney & Rolfes, 1993a). The importance of zinc during early infancy was first documented in 1970 (Shaw, 1979). Hambidge, Walravens, Casey, Brown & Bender (1979) suggested that zinc status in young children and infants may be suboptimal. Hambidge and Silverman (1973) reported an 18 months old infant with failure to thrive, poor appetite and pica; these signs appeared at 6 months of age. Treatment with zinc rapidly abolished these problems. In very young infants this deficiency can be caused by an exceptionally low concentration of zinc in mothers milk (Kuramoto, Igarashi &

Tagami, 1991). There are several recent case reports of zinc deficiency in premature infants receiving breast milk shown to be low in zinc (Aggett et al., 1980; Blom, Jameson, Krook, Larsson-Stymne & Waranne, 1980; Zimmerman et al, 1982). It has been proposed that the mode of feeding may have a major impact on the zinc status of infants. The plasma zinc concentration is higher in breast fed infants than in formula fed infants. This observation is surprising as formulas, usually based on cow's milk, contain zinc in amounts equal to or higher than that of human milk. Thus, it has been suggested that the bioavailability of zinc from mother's milk is superior to that of zinc in cow's milk. The superior availability of zinc in human milk is believed to be due to its much higher contents of a low molecular weight zinc binding ligand (ZBL) (Lonnerdal, Stanislowski & Hurley, 1980). Acrodermatitis entropathica (patients unable to absorb sufficient zinc from any other food), does not manifest during breast feeding. This suggests that despite a lower zinc concentration, human milk is superior to cow milk in providing bioavailable zinc (Casey, Walkavens & Hambidge, 1981; Sandstrom, Cederblad & Lonnerdal, 1983).

Sandstrom et al. (1983) investigated the absorption of zinc from human milk, cow milk, cow milk formula (Whey – adjusted), and soy protein formula in 16 day old suckling rats as these fluids can comprise the majority of an infant's diet. The results showed that the zinc bioavailability was 28% from human milk, 24% from whey adjusted cow milk formula, 15% from cow's milk and 10% from soy formula. The study also showed that the rat pup model might provide a rapid, inexpensive, and sensitive method to assay bioavailability of zinc from infant food.

There is a greater concern for zinc deficiency in the developing world because of the lack of animal products like meat, fish in the diet. Use of low zinc starchy roots and tubers as food staples, and intakes of phytate from plant-based foods are sufficient to negatively affect the bioavailability of dietary zinc (Gibson, 1994). It may be partially responsible for impaired neuro cognitive development that is associated with poor growth in young children (Hambidge, 1988). There is growing evidence that zinc deficiency contributes to some of the major causes of morbidity among young children in the developing world and has been well illustrated by recent studies in New Delhi, India (Sazawal, Black & Bhan, 1996). Results of zinc supplementation have included reductions in the prevalence, severity, and duration of diarrhea and have important implications for diarrhea-related morbidity and mortality.

In the US an increasing number of infants are being fed soy formulas because of gastrointestinal problems and for other reasons. Although soy formulas have been designed to meet the nutrient requirements of infants, phytate in soy lowers the zinc absorption (Lonnerdal et al., 1999). Reducing the phytate content of soy formulas by the 2 step precipitation method significantly enhanced the zinc absorption in infant rheus monkeys. This method of reducing the phytate content of soy formula is relatively costly and has not yet been applied on a commercial scale.

Foods that are high in protein content are high in zinc, such as shellfish, meats, liver, and eggs. Whole grain products, legumes, and nuts are good sources of zinc if large quantities are eaten. In a typical US infant diet phytate intake from grains is not enough to impair zinc absorption (Whitney & Rolfes, 1993a). In India a typical infant diet consists of whole grain cereals, legumes, fruits, and milk. The diet contains relatively little or no meat, fish, or other animal products high in zinc due to socio-economic reasons or religious beliefs. The dependence on plant food (grains and cereals) for the provision of zinc suggests low availability and potential chelation of zinc by phytate, thus compromising the zinc supply (Paul, Bates, Prentice, Day & Tsuchiya, 1998). Both naturally occurring phytate and sodium phytate added to phytate have been shown to inhibit zinc absorption to the same extent (Fairweather-Trait & Caprez, 1982). The greatest impact of phytic acid on human nutrition is its reduction of zinc bioavailability. Numerous studies have implicated phytic acid as a causative factor in poor zinc absorption from plant foods (O'Dell, 1969; Likuski & Forbes, 1965). Using a range of dietary minerals, Maddaiah, Krunick & Reid (1964) and Vohra et al. (1965), found that, in vitro, zinc formed the most stable complex with phytate and calcium has been shown to form the least stable complex with phytate. Zinc is known to interact with numerous dietary components, which influence its availability. These include certain other minerals like calcium, Vitamin D, phosphate, and chelating agents such as amino and organic acids. Both the level and source of dietary protein are important determinants of zinc availability.

Calcium

Ninety nine percent of the body's calcium is located in the bones, where it plays two roles. First it is an integral part of bone structure, and second it serves as a calcium bank offering a readily available source of minerals to the body fluids.

Rickets is a common syndrome resulting in malnutrition in infants and children and is an impairment of faulty calcification resulting in misshaped bones (bowing of legs), retarded growth, enlargement of ends of long bones (knees and wrists) and deformities of ribs. Historically rickets grew in importance as the industrial revolution brought about migration of laborers from country side to city. Smoke from tens and thousands of coal fires combined with cloud, fog and the dimmed light of northern latitudes deprived infants and children of sunlight necessary for vitamin D synthesis. Epidemics of rickets resulted. There is convincing evidence that lack of sunlight could cause rickets and that exposure to sunlight was curative. It remains prevalent in the subtropics including the middle east, northern India, and the northern tier of Africa.

Rickets, long prevalent in India, has been the subject of many studies. Religious and folk traditions continue to be important determents of life patterns in developing countries. This is true especially of the moslem group. Loomis (1970) reported that the incidence of rickets in India among children from prosperous moslem families was seven times the rate observed in hindu families of similar economic status. Deprivation of sunlight among the moslems explains the difference. Wilson and Widdowson (1942) concluded from studies of India that vitamin D deficiency alone was unlikely to produce rickets in infants. Besides the diet deficiency of calcium and vitamin D, other factors also influence the production of rickets like the action of antimetabolities, dietary phytate and fiber.

The bioavailability of dietary nutrients to a large extent is determined by a net effect of inhibitors and promoters. Utilization of calcium from milk is high (Heaney &

Weaver, 1989). The occurrence of phytic acid as mixed calcium and magnesium salt in grains was first reported by Pfeffer (1872).

Phytates are present in large amounts in most plant foods. These plant foods are the major source of nutrients for infants in India and other developing countries. Phytate is identified as an anti-nutritional factor impairing calcium utilization; phytate as a component of cereals is most responsible for decreased deposition of calcium in bones. The action of phytate was not reversed by vitamin D. Phytate added to the diets of infants and children in amounts calculated to combine with 50 to 100% of the dietary calcium caused a considerable decrease in calcium absorption (Jorgensen, Anderson & Nielsen, 1946). The rickets producing effects of phytate are not fully understood, but appear to be due in part to the poor availability of phytate phosphorus and partly to the ability of phytate to bind calcium to form an insoluble complex from which calcium is unavailable for absorption. These two actions also result in an elevated vitamin D requirement, which promotes the absorption of calcium. The most important source of this vitamin is biosynthesis in the skin through sunlight. Very little is supplied by unfortified diets in developing countries. Rajalakshmi (1976) observed that calcium supplements to diets of Indian school children promoted bone development more effectively than did vitamin D, indicating that the vitamin failed to overcome the retardant effect of phytate.

In many parts of the world, plant foods are the most important dietary sources of calcium (Singh, Sharma & Sur, 1969; Walker, Walker & Wadvalla, 1968). The availability of calcium from cereals and vegetables is lower than that from milk and other animal foods because of their generally lower protein contents and also because they often contain an abundance of substances which greatly inhibit the absorption of calcium as well as many other minerals. Much of the calcium in plant foods is found in combination with these inhibitors, which include oxalic and phytic acid, and fiber.

The Recommended Dietary Allowance (RDA) are a set of nutrient standards established by the Committee on Dietary Allowances. The recommended dietary allowances for calcium, zinc, and iron for infants are shown in Table 2.

Molar Ratios

To determine the bioavailability of minerals in phytate containing foods for humans, it was essential that a tool be devised for predicting consequences of phytate/mineral interactions. Since human studies are expensive and difficult to conduct, Oberleas (1975) suggested that a reliable indication of zinc availability from phytate rich foods might be obtained from the phytate and zinc contents expressed as the molar ratio phytate/zinc. In support of this proposal he demonstrated reduced growth rates in rats fed soybean-based diets containing phytate/zinc values from 3.2:1 to 495.5:1. However, in their research only a relatively small number of widely varying phytate/zinc values were used and some of the diets were deficient in zinc even without consideration of their phytate contents.

Davies and Olpin (1979) further refined the ratio. They reported that dietary phytate in rat feeding trials caused significant reductions in growth rates, plasma zinc concentrations, and hair zinc concentration, as well as graying of the coat at phytate/zinc molar ratios as low as 10. They concluded that phytate/zinc molar ratios could be used as an indicator of zinc availability from phytate rich food and phytate/zinc ratio of 10 or less

Table 2: Recommended Dietary Allowance for Infants

Infant Age	Calcium (mg)	Iron (mg)	Zinc (mg)
Birth – 6 months	400	6.0	5
6 months – 1 year	600	10	5

Note: From Understanding Nutrition By Eleanor Noss Whitney & Sharon Rady Rolfes, 1993, St. Paul:West Publishing Company, p.1.

were associated with adequate zinc status and ratios above 20 may be associated with clinical or chemical evidence of zinc deficiency.

Oberleas and Harland in 1981, published phytate and zinc contents in several foods. The phytate and zinc values were used to calculate phytate/zinc molar ratios. The phytate/zinc molar ratios of foods obtained in their study were estimated for relative risk of having an inadequate intake of zinc based on the ratio of 10 or less being with adequate zinc bioavailability. According to Oberleas and Harland (1981), the phytate/zinc molar ratios of 10 or less are probably free of significant effects of phytate on zinc. These ratios can be used in planning menus to select the combinations of foods that will supply the most available zinc to the daily diet. The phytate/zinc molar ratios of foods has been fairly well documented (Oberleas & Prasad, 1976; Harland, 1978;Oberleas,1983a, b; Harland & Oberleas, 1985).

After the proposal for use of a phytate/zinc molar ratio, Morris and Ellis (1985) proposed the application of phytate/calcium molar ratios to humans, suggesting that dietary intakes with phytate/calcium molar ratios greater than 0.2 may be under the risk of calcium deficiency.

Commercial Baby Foods in India and their Ingredients

This study involves five commercially prepared baby foods used for infants in India. These foods are manufactured in India as indicated in Table 3. The ingredients used in the baby food are rice, wheat, mung bean, milk, and oranges.

At about six months of age, the quantity of breast milk supplied by the mother is insufficient to meet the energy and nutrient requirements of the growing infant.

Introduction of semi-solid foods to the diet, preferably with relatively high energy and nutrient densities, is important to prevent malnutrition. Such foods must not only be safe, acceptable to taste, and easily affordable, but also nutritious.

The popular baby foods/weaning foods available in the markets of India are developed by blending cereals and legumes with or without skim milk (Desikachar, 1980). Mung bean is normally used in homes for preparing rice-mung bean weaning foods (Roxas, Intengann, & Juliano, 1976). Inclusion of various legumes in the diet has been shown to promote the growth of under weight children (Adsule, Lawande, & Kadam, 1984). Commercial infant cereals are usually precooked, roller dried cereal flours that are composed primarily of cereal with a low extraction rate, such as wheat, and rice, and are served with milk. These cereals are usually fortified with minerals (Davidsson et al., 1997). Unfortunately the phytic acid, dietary fiber, and polyphenol contents of many plant-based foods are high. These dietary components are known to inhibit the absorption of some major and trace minerals (Gibson et al., 1998). In India several types of commercially prepared foods have been developed to fulfill the nutritional needs of infants and to satisfy the requirements of caregivers for convenience and safety. Some of the baby foods manufactured in India are presented in Table 3. These baby foods are not only manufactured and used in India, but are also used in Saudi Arabia (Al-Othman, Khan & Kanhan, 1997), and Pakistan (Khan & Kissana, 1985).

Foods mentioned in Table 3 are all processed, precooked, dry cereals, and are therefore low in moisture. These baby foods are fortified with vitamins and minerals. Baby foods are important in supplementing the diets of infants whose needs for nutrients

Table 3. Types of Indian Baby Foods

Baby Food	Manufacturer
Nestrum Rice	Nestle India Ltd.
Cerelac Wheat	Nestle India Ltd.
Cerelac Wheat – Orange	Nestle India Ltd.
Farex Wheat – Mung Bean	Hindustan Foods Ltd. & Heinz India
Dexolac Rice	Wockhardt India Ltd.

are not met by human milk, infant formula or other milk (Purvis & Bartholmey, 1988). In order to reduce the incidence of allergy arising from an unnecessarily complex diet, single grain cereals such as rice, oat meal, and barley are generally recommended as starting cereals. Mixed cereal based on three or more grains (oat, corn, wheat, rice, soy) provides a greater variety to older infants. High protein cereals based on soy flour and wheat flour offer low cost and high protein alternatives in addition to variety. Cereals with fruit appeal to infants with those flavor preferences and also provides a rich source of iron (Purvis & Bartholmey, 1988).

Rice

In India, rice in the form of rice flour or granulated rice is used in the formulation of baby foods, particularly cereals and combinations of legumes, fruits, and vegetables. Precooked infant rice cereals are frequently prescribed as the infant's first solid food. These cereals are excellent vehicles for the introduction of iron and other essential minerals and vitamins into the infant's diet. Rice cereal, because it is considered to be hypoallergic, is usually the cereal of first choice for infants even in the United States. Each baby food manufacturer has its own formulation and process for manufacturing rice cereal. Since rice cereal is an easily digested first cereal, it is very important that the product be acceptable to both baby and parents (Kelly, 1985).

Nutritive Value of Rice

Rice has the lowest protein content among cereals. It is also low in fiber and fat content. The amount of B vitamin, iron, and zinc content is low. Iron and zinc bioavailability in rice is low as is true with most vegetables or plant sources. The mineral composition of rice grain depends considerably on the availability of soil nutrients during crop growth. The nutritional value of rice is shown in Table 4. Several antinutritional factors are concentrated in the bran portion of the rice. The phytin content of rice bran is higher than that of wheat bran (Thompson & Weber, 1981). Most of the antinutritional factors are protein in nature except for phytin. Phytin is located in the aleurone portion of rice as potassium magnesium salt. Its phosphate groups can readily complex with cations such as calcium, zinc, iron and protein.

Wheat

Wheat and wheat containing foods are a major staple and source of calories in the diets of people in India and many other countries. Although wheat foods are often seen mainly as a source of carbohydrate, wheat foods are also a substantive source of protein, vitamins, and minerals when consumed as a major component of the diet. In recent years publicity has been given to the nutritional benefits of low-fat, high fiber diets. This has heralded an uplifting of the status of cereal products throughout the world, and they are now regarded as essential components of a healthy diet. Wheat flour is the major wheat product, used for making baby foods, and a variety of palatable products. The wheat kernel has three parts: the bran, the endosperm, and the germ (embryo). The bran has a protective function, and the endosperm is a food storage site for the developing plant (Orth & Schellenberger, 1988).

The distribution of the nutrients within the wheat kernel is typical of that of many cereals. Although the endosperm consists mainly of starch, since it makes up the major portion of the wheat kernel, a significant proportion of many minerals and vitamins in

Table 4: Nutritional Values of Wheat and Rice*

	Wheat	Rice
Proximate Composition		
Protein(g)	10.5	7.5
Fat (g)	1.9	1.8
Carbohydrate + Fiber (g)	74	77
Minerals		
Calcium (mg)	35	15
Iron (mg)	3.9	1.4
Vitamins		
Thiamine (mg)	0.38	0.33
Riboflavin (mg)	0.08	0.05
Niacin (mg)	4.3	4.6

* based on 100 grams, edible portion

Note: From Wheat Chemistry and Technology by Y. Pomeranz. 1988, St. Paul:American Association of Cereal Chemists, Inc., p. 12.

wheat is located in the endosperm. Nutrients are generally found in highest concentration in the germ or embryo and in the aleurone cells surrounding the starchy endosperm. Aleurone cells are rich sources of minerals, many of the B vitamins, protein, and phytic acid. Nutrients of wheat and their location in the kernel are shown in Table 5. Significant quantities of the minerals and vitamins are lost when whole wheat is milled to produce white, endosperm flour because the outer layer of bran is removed along with the aleurone cells and the germ (Betschart, 1988).

Wheat is a major component of most diets of the world because of its agronomic adaptability, ease of storage, nutritional goodness, and the ability of its flour to produce a variety of palatable, interesting, and satisfying foods (Moss, 1973).

Nutritive Value of Wheat

All cereal grains are high in carbohydrate because the main component of the endosperm is starch, which is embedded in a protein matrix. The composition of wheat varies depending on the variety and growing conditions, including rainfall, soil, temperature and the fertilizers used. Wheat contains considerably more protein, on average, than other cereals. This factor accounts for the better nutritional status of wheat eating people.

As a source of protein, cereals are characterized by a low lysine content relative to the reference protein of the Food and Agriculture Organization and The World Health Organization (FAO/WHO, 1984). Lysine is thus the limiting amino acid. However, foods based on staple cereal supplemented with small quantities of legumes, nuts, and milk are nutritionally adequate for protein when energy requirements are met. The fat content of

Table 5: Wheat Nutrients and Their Location in Kernel

Fraction	Nutrients Present
Bran	Dietary fiber, potassium, phosphorus, magnesium, calcium
Aleurone	Niacin, minerals (especially phosphorous), phytic acid
Endosperm	Starch, protein. Pantothenic acid, riboflavin, minerals
Germ	
Embryo	Fats and lipids, sugars
Scutellum	B vitamins (especially thiamine), phosphorous

Note: From Wheat Chemistry and Technology by Y. Pomeranz. 1988, St. Paul:American Association of Cereal Chemists, Inc., p. 11.

wheat is slightly higher than rice. Cereals are major contributors of dietary fiber, B vitamins, and supply a variety of trace minerals. The nutritional value of wheat is shown in Table 4.

Phytic acid contains as much as 70-75% of the total phosphorous content of wheat (Booth, Carter, Jones & Moran, 1941). It forms insoluble salts with various metals and can interfere with the assimilation of zinc, calcium, and iron in human and animal nutrition, particularly when the fibrous part of wheat is an important part of the diet (Kent-Jones & Amos, 1957).

Mung Bean

Vigna Radiata (L.) Wilczek is commonly called green gram, golden gram or mung bean. Food legumes are relatively inexpensive sources of nutrients in developing countries. Consumption of legumes in India is known to be very high. Together with staple cereals like wheat, rice, millet, and mung beans, pigeon peas, and chick peas are not only very popular with adults, but are also well liked by infants and children. Many deep fried products and weaning foods are prepared by using legume flour. This indicates various modes of utilization of legumes as human food (Salunkhe & Kadam, 1989b).

Mung bean has been cultivated in India since ancient times and is still an important source of food for humans. In India, the mung bean ranks third in production next to the chickpea, and pigeon pea and is extensively grown on all types of soils and under varying climatic conditions. It has been reported that mung bean is an excellent source of protein with higher digestibility (Bressani, Elias & Altschul, 1974; Salunkhe, 1982). Mung bean is often fed to babies with cereals, consumed by the elderly or used when ending a long fasting period (Salunkhe & Kadam, 1989b).

Nutritive Value of Mung Bean

Mung bean has three major components, namely seed coat, cotyledon, and embryo. The nutrients are distributed evenly in different anatomical parts of the mung bean. The embryo has the highest content of protein and lipids while starch and crude fiber are concentrated in the cotyledons and seed coats respectively (Singh, Ingh & Sikka, 1968). Due to the relatively high content of lysine in mung bean, which is deficient in cereals, the combination of cereals and mung bean produces a good balance of amino acids since the cereal supplies adequate methoionine (Chatterjee & Abrol, 1975). Minerals such as calcium, phosphorous, sodium, iron, and potassium are commonly found in mung bean. Calcium and iron are mostly concentrated in the seed coat, and phosphorous is mostly found in the embryo.

The proximate composition and mineral content of mung bean are shown in Table 6. Food legumes are known to contain several antinutritional factors such as the trypsin inhibitor, polyphenols, cyanogenic compounds, goitrogens, and phytates. Phytic acid (phytate) is widely present not only in cereals, but also in legume seeds (O'Dell, 1979). It is a chelating agent for cations (Cosgrove, 1966; Sobolev, 1966). Phytate rapidly accumulates in seeds during the ripening period accompanied by other storage substances such as starch and lipids (Abernathy, Paulson & Ellis, 1973; Nahapetian & Bassiri, 1976). The accumulation site of the phytic acid in seeds is in the globoids (Tanaka, Yoshida & Kasai, 1974; Lott & Buttrose, 1978). The amount of phytic acid varies from

Table 6. Nutritional Values of Mung Bean*

	Mung Bean
Proximate Composition	
Protein(g)	22.9
Fat (g)	1.2
Carbohydrate + Fiber (g)	66.2
Minerals	
Calcium (mg)	105
Iron (mg)	7
Vitamins	
Thiamine (mg)	0.53
Riboflavin (mg)	0.26
Niacin (mg)	2.5

* based on 100 grams, edible portion

Note: From Handbook of World food Legumes: Nutritional Chemistry, Processing Technology, and Utilization Volume II by D. K. Salunkhe, & S. S. Kadam, 1989, Boca Raton, FL:CRC Press, Inc., p.66.

0.40 to 2.06% in legumes (Reddy, Sathe & Salunkhe, 1982). In mung bean the amount of phytic acid content is 0.66% (Kumar, Venkataraman, Jaya & Krishnamurthy, 1978). In many cases, the phytic acid content varies depending on the variety or cultivar, climatic conditions, locations, irrigation conditions, type of soil, and the year during which they are grown (Salunkhe & Kadam, 1989a).

Oranges

More than any other fruit, oranges are associated with and valued for their vitamin C content (Margen, 1992). Although the iron content of oranges is low, they may help to improve the iron bioavailability of the diet as a whole. Oranges contain ascorbic acid, citric acid, and other organic acids, which increase iron absorption both by reducing ferric iron to ferrous iron and by enhancing iron solubility. Thus oranges added to infant foods are likely to enhance the iron status of infants (Gillooly et al., 1984). Oberleas (1973) reported that oranges contain no phytates.

Phytate as an Anticarcinogen

Inositol hexaphosphate (IP6) is synthesized from inositol (Ins). (Holub, 1992). Adults consume about 1 gram of inositol per day from both animal and plant sources. Several mammalian tissues can synthesize about 4g/day: testis, mammary gland, brain, liver and kidney. It is synthesized from D-glucose. Although plants routinely synthesize IP6, the evidence that humans do is sparse. Humans cannot degrade IP6, whereas yeast, fungi, plants, birds and some animals can (Harland & Morris, 1995).

Dietary factors appear to have a strong influence on the development of cancer of the large intestine. Cereals and legumes by virtue of their higher phytate content appear to be most protective. By complexing iron, phytate may bring about a favorable reduction of hydroxyl radicals in the colon (Graf & Eaton, 1993). Most of the historical literature concerning phytate emphasizes its mineral-chelating properties, to the detriment of animal and human health. Pretlow, O'Riordan, Somich, Amini & Pretlow (1992) have suggested that phytate added at a 2% level to drinking water of rats may be used as an intermediate biomarker for rat tumor incidence. In a 36-week study, rats were injected with azoxymethane to induce colon cancer. Beginning stages appeared as aberrant crypts in unembedded segments of colon tissue. Untreated, these would progress to tumors in a dose-response fashion. The higher the incidence of aberrant crypts, the greater the tumor formation. Adding phytate to the drinking water reduced the number and depth of the colonic crypts when rats were examined at 12 and at 36 weeks. The incidence of tumors in rats denied phytate water was 83% compared with 25% in rats drinking phytate water. This study also showed that phytate in the drinking water administered after the azoxymethane injection could not prevent the formation of aberrant crypts, but it could prevent the next progressive step of tumor formation.

CHAPTER 2

JOURNAL ARTICLE

Authors' Title Page

PHYTATE AND MINERALS IN INDIAN BABY FOODS

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Summary

The objective of this study was to analyze five commercially available Indian baby foods for phytate, calcium, iron, and zinc. Molar ratios of phytate/calcium, phytate/iron, phytate/zinc were calculated to determined the bioavailability of these essential minerals. The baby foods used in this study were based on either wheat or rice. Phytate, calcium, iron, and zinc content were determined using AOAC method.

The phytate content (mg/100g) ranged from 1.22 (Dexolac) to 1.97 (Farex). Iron content (mg/100gm) ranged from 8.0 (Cerelac Wheat) to 10.0 (Farex). Zinc content (mg/100gm) ranged from 2.8 (Nestrum) to 8.5 (Cerelac Wheat) and calcium content (mg/100gm) ranged from 97.5 (Nestrum) to 333.0 (Dexolac). Molar ratios of phytate/iron ranged from 0.006 (Nestrum) to 0.018 (Cerelac Wheat). Molar ratios of phytate/zinc ranged from 0.019 (Cerelac Wheat) to 0.045 (Nestrum). Molar ratios of phytate/calcium ranged from 0.0002 (Dexolac) to 0.0008 (Nestrum). Molar ratios of phytate/zinc and phytate/calcium were well below the published value associated with adequate zinc and calcium bioavailability of 10 and 0.2 respectively. The results showed that, the phytate content of the baby foods was unlikely to inhibit bioavailability of zinc and calcium. As phytate concentration was low in all the baby foods analyzed, it was concluded that iron was also bioavailable.

Keywords: Phytate, Baby Foods, Calcium, Iron, Zinc, Molar Ratio, Bioavailability

Introduction

The best food for infants is breast milk. In the first three or four months of an infant's life, a period of rapid growth, most infants thrive well on their mother's milk alone. However, by five to six months of age, the supply of energy and some nutrients from breast milk are no longer adequate to meet the growth requirement. The infant needs other foods besides milk which supply energy, protein, minerals, and other nutrients. It has been reported that early weaning is a common practice in India and other developed countries where socio-economic levels are high (Dodd & Datta, 1988). In a study conducted by Gibson et al. (1998), the complementary foods used in developing countries were reported to be mainly from cereals or starchy roots and tubers. They are prepared as thin gruels. The energy and nutrient contents of these gruels are low. It is further exacerbated if the infants receive very few feedings per day.

Malnutrition is widespread in India and many developing countries; the condition is particularly serious in children below 3 years of age. It is estimated that between 30% to 50% of the children in this group are malnourished (Ashturkar et al., 1992). Malnutrition in many regions may not be due to energy and protein deficiency alone. It may be a cumulative affect of many factors such as large families, poverty, ignorance, deficient diets, infection, or poor absorption of minerals (Sehgal et al., 1989).

In India cereals and legumes serve as the main and economic source of nutrients such as carbohydrates, protein, vitamins, and minerals in an infant's diet. The combination of cereals and legumes provide a good balance of amino acids (Salunkhe &

Kadam, 1989). In India several types of commercially prepared foods have been developed to fulfill the nutritional needs of infants. The popular baby foods available are developed by blending cereals, legumes, and fruits, with or without skim milk (Desikachar, 1980). The baby foods Cerelac Wheat, Cerelac Wheat-Orange, Farex, Nestrum, and Dexolac used in this study are mainly based on wheat, rice, mung bean, oranges, and milk. The plant-based foods contain a major portion of phosphorous as phytate phosphorous. Phytate, the inositol hexaphosphate, is a complex salt, which occurs naturally in plant foods (Harland & Morris, 1995). The phytate molecule is negatively charged and is known to complex or chelate with metallic ions of minerals like zinc, calcium, and iron forming insoluble compounds and interfering with mineral utilization (Gustafsson, & Sandberg, 1995). Formation of insoluble compounds results in a decrease in mineral bioavailability, particularly in infants.

Iron deficiency is a worldwide problem affecting a large number of people in western communities as well as in underdeveloped countries. It is more prevalent in infants, and is usually the result of low bioavailability of dietary iron (Fox et al., 1998). Infants and children have higher needs for zinc because they are growing rapidly and synthesizing many zinc containing proteins (Whitney & Rolfes, 1993). Calcium malnutrition in the form of rickets affects a wide number of infants.

Many studies have suggested that molar ratios of phytate/zinc, phytate/calcium, and phytate/iron may provide useful indexes of bioavailability of minerals (Ellis et al., 1987). Reliable indications of zinc availability from phytate rich food might be obtained from the phytate and zinc contents expressed as the molar ratio of phytate/zinc (Davis & Olpin, 1979).

The aim of the present study was to determine the phytate, calcium, iron, and zinc contents of five commercially available Indian baby foods. Molar ratios of phytate/calcium, phytate/iron, and phytate/zinc were determined to estimate bioavailability of calcium, iron, and zinc in the five baby foods analyzed.

Materials and Methods

Five commercially available baby food products were obtained from a local food store in New Delhi, India. These baby foods are popular and commonly used in urban areas of India. All of these baby food cereal products were in dehydrated coarse powder form. Some of these baby foods were sealed in aluminum pouches and packed in cardboard cartons and some foods were directly sealed in aluminum tins. Each carton or tin of the baby food has information on: the instructions for preparation, ingredients, nutrient composition, date of manufacture, and date of expiration. All of the baby foods are precooked ready to eat cereals, and one just needs to add water. The description of these baby foods is as follows:

Cerelac Wheat

Cerelac Wheat contains wheat flour and partially skimmed milk. This precooked food has been designed to be easily digested by babies and it is recommended to begin use at 4 months of age. This food has been fortified with vitamins and minerals.

Cerelac Wheat- Orange

In addition to wheat flour and partially skimmed milk this food contains orange concentrate which makes it flavorful. Since it is precooked, it is easily digested by babies. This food is recommended for infants ages 4 months and above. It has been fortified with vitamins and minerals.

Farex

Farex contains wheat flour, milk solids, and mung bean. This food is recommended for infants after 5 months of age. Farex is scientifically formulated to provide a balanced mix of essential nutrients to suit the baby's delicate digestive system. This food already contains milk, and one just needs to add water. All of the Farex variants are precooked (making them easy to digest) and do not contain any artificial preservatives, colors or flavors.

Nestrum

This is a rice-based cereal made from quality rice flour. This food contains no milk or milk products. Nestrum has been precooked and fortified with vitamins, iron-salts, and calcium carbonate to suit the baby's tender digestive system. This food can be used for infants beginning at 4 months of age.

Dexolac

This food contains extruded rice flour and partially skimmed milk solids. It is recommended to begin at 4 months of age. This food is fortified with mineral salts and vitamins.

Phytate Determination

Phytate was analyzed according to AOAC official method (1995), using the anion exchange separation method. Two gram samples from each baby food were accurately weighed and extracted with 40 ml of 2.4% hydrochloric acid for 3 hours in a mechanical shaker at room temperature. After 3 hours the sample was filtered using whatman no. 1 filter paper and the filtrate was used for phytate analysis. To avoid hydrolysis from mold or bacterial enzymes, the filtrate was refrigerated no more than one week before analysis.

Anion exchange resin columns (0.7 x 15 cm) were prepared using water slurry of 0.5 gram of AG1 – X4, 100-200 mesh, resin in chloride form. After the resin bed was formed, the columns were washed with 15ml of 0.7M sodium chloride. The column was eluted with 15ml of 0.7M sodium chloride to ensure that the resin was in the chloride form and then was washed with 15ml of distilled water until the eluate was salt free.

First the blank was passed into the prepared resin column consisting of 1ml of 2.4% hydrochloric acid with 1 ml of disodium ethylene diamine tetra acetate – sodium hydrochloride reagent, and was diluted to 25ml with distilled water and eluted by gravity. Second, the 5.0 ml of filtrate sample was pipetted to 1 ml of disodium ethylene diamine tetra acetate – sodium hydrochloride reagent, and was diluted to 25 ml with distilled water. The extract was applied to the column by mixing and transferring quantitatively. The eluate was discarded, and the resin was washed with 15 ml of distilled water. The contents of the column were then eluated with 15 ml of 0.1M sodium chloride to eluate any inorganic phosphates and then were discarded. The phytate anion was eluated with 45ml of 1.4M sodium chloride and collected.

The eluates were digested in digestion tubes with 0.5ml sulfuric acid and 3ml of nitric acid with some glass beads under the hood on micro-Kjeldahl rack over medium heat. The digestion was carried out until active boiling stopped(25 – 30 min.). The end point was reached when a cloud of thick yellow vapors filled the neck of the tube. Contents were heated 5 minutes more on medium heat. When the flask was cooled about 10ml of distilled water was added. Then it was heated for 10 minutes on a low temperature setting to dissolve the salt and hydrolyze any pyro phosphate that may have formed during digestion. Digestion time is critical because incompletely digested products will not react with color reagents and over digestion will cause sublimation of the phosphorous (Jones, 1949). The solutions were cooled and transferred quantitatively to 50ml volumetric flasks. Two ml of molybdate solution and 1ml of sulfonic acid reagent (reducing agent) were added to the digested extract. Finally with the distilled water, a volume of 50ml was made. After 15 minutes the absorbance was read in the spectrometer at 640 nm. To detect the amount of phosphorus 1.6, 4.8, and 8.0 ppm of phosphorus standard solution was pipetted into 50ml volumetric flasks. To the flask, 2ml molybdate solution and 1.0ml of sulphonic acid were added and made to the volume of 50ml with distilled water. The phosphate standards were prepared from phosphorous standard solutions (1.0, 3.0, & 5.0ml) and were read after 15 minutes in the spectrophotometer at 640nm. The amount of phosphorus obtained from the standard phosphorus curve was converted to phytate using the following formula.

Phytate, mg/g sample = "mean K" x A x 20/(0.282 x 1000)

Where A = absorbance; "mean K" = standard P(μg)/A/n(standards);

phytate = 28.2%P.

Mineral Analysis

Calcium, iron, and zinc were determined by the atomic absorption spectrophotometric method (AOAC, 1995). The residue obtained from ash determination in proximate analysis was dissolved in 1N HCL and transferred quantitatively into a 25 ml volumetric flask before diluting to the volume; an amount of lanthanum (0.125 gms) equivalent to 0.5% in the final solution was added. The mixture was then made up to a volume with 1N HCL. Appropriate dilutions of the final solution were made in order for the absorption to fall within the absorption values of standard solutions of calcium, iron, and zinc.

Concentrations of calcium, iron, and zinc in the five commercially available baby foods were determined from the standard curves prepared for each mineral. Three standard solutions, each containing a mixture of a known amount of calcium, iron, and zinc, were prepared from their corresponding stock solutions. Standard solution 1 contained 0.8, 0.8, and 0.20 ppm of calcium, iron, and zinc respectively. Standard solution 2 contained 2.5, 2.5, and 0.5 ppm of calcium, iron, and zinc respectively, and standard solution 3 contained 5.0, 5.0, and 1.0 ppm of calcium, iron, and zinc respectively. To each of these standard solutions was added lanthanum to a concentration of 0.5%(w/v).

Atomic absorption of calcium, iron, and zinc in diluted ash solutions and standard solutions were read at 422.7, 372.0, and 213.9 nm, respectively. The concentration of the three minerals were obtained from the standard curves.

Molar Ratios of phytate/minerals

To determine the bioavailability of iron, zinc, and calcium in Indian baby foods, molar ratios of phytate/iron, phytate/zinc, and phytate/calcium were calculated using contents of phytate, iron, zinc, and calcium obtained in the present study. Molar ratios are the number of moles of phytate to one mole of the mineral.

Proximate Analysis

Moisture, crude fat, crude protein, and ash contents of the five commercially prepared baby foods were determined by using the standard procedures of the Association of Official Analytical Chemists (1990,1995). The total carbohydrate content of the five baby foods were determined by difference, subtracting the percentage of moisture, crude fat, crude protein, and ash from 100%.

Results and Discussion

Phytate Content

The phytate contents (mg/100g) of five commercially prepared Indian baby foods are shown in Table A. The phytate content of the baby foods ranged from 1.22 in Dexolac Rice to 1.97 in Farex (Wheat & Mung Bean). Farex had the highest phytate content and Dexolac had the lowest phytate content. There is not much variation in the phytate content of baby foods; the values are close to each other. It appears that baby foods prepared from wheat have a slightly higher level of phytate than rice-based baby foods. The phytate contents of wheat-based baby foods, Cerelac Wheat and Cerelac Wheat – Orange, are close because citrus fruit oranges does not contain phytate (Harland & Morris, 1995). Baby food Farex contains the highest level of phytate (1.97 mg/100gm) compared to the rest of the baby foods, as it has mung bean in addition to the wheat. Raw mung bean has a level of 0.66% phytic acid (Kumar et al., 1978). However, the small variability of phytate content among the rice based baby foods Nestrum and Dexolac can be related to use of different methods and condition of processing done by the manufacturer.

Many studies have reported the phytate contents of raw foods. Very few are available on precooked baby foods, hence making it difficult to compare the analyzed value for phytate in baby foods in the present study. The baby foods used in this study were commercially prepared, precooked, and were fortified with minerals and vitamins. The kind of processing and variety of cereals and flour (wheat and rice) used in the food were not mentioned on the label. Due to the limited studies on phytate content of Indian baby foods and limited knowledge of the processing, and the variety of cereal used in the present study, the phytate content of Indian baby foods were compared to the phytate content of raw ingredients. The analyzed value for phytate content of wheat and rice used in the baby foods were much lower than the phytate content of raw wheat and rice. During processing, phytate present in the aleurone, germ, and bran portion of these cereals are removed. It is possible that during cooking a considerable amount of phytate in foods is reduced (Hallberg et al., 1989). The degradation of the inositol hexaphosphate results in lower molecular forms such as tri- or tetra phosphates, thereby lowering the content of phytate (Sandberg et al., 1988).

As studies indicate that phytate is known for its high content in plant-based foods like cereals and legumes, commercial infant cereals are produced from low extraction rate wheat and rice cereal flours with a relatively low content of phytic acid, a strong inhibitor of mineral bioavailability. These foods are usually fortified with minerals because processing also removes essential minerals and vitamins.

In the present study there is a lack of knowledge of the kind of extraction performed on the cereals. Since it is stated on the baby food containers that the foods are formulated to suit the delicate digestive system of babies, an assumption was made that the cereal flours used are of low extraction rate and most of the phytate was degraded during processing.

The values obtained for phytate content in the present study (1.22 – 1.97 mg/100g) are much lower than the findings of Gibson et al. (1998) in the determination of phytate content of complementary foods for infants used in developing countries. Their values of phytate content in infant food ranged from 5 – 203 mg/100g. However, results of the study by Gibson et al. cannot be equally compared to results of this study due to the fact that their foods were prepared from unrefined cereal flours like maize. The high level of phytate obtained in their study was due to the use of unrefined maize flour, which contains most of its phytic acid (90%) in the germ. The lower values for phytate content in their study were in wheat-based complementary food, which consisted of Sago flour, brown sugar, oil, and water.

The values obtained for phytate content in this study were also compared with the findings of Paul et. al. (1998) in the determination of phytate intake of rural Gambian infants. Their analyzed value for phytate ranged from (40-240 mg/100g). The foods were based on rice, millet, sorghum, and groundnut. The researchers related the high amount of phytate obtained in their study to the analytical method, Fe-precipitation, which not only identifies inositol hexaphosphate but also the lower phosphate esters of inositol, although these esters have less adverse effect on mineral availability (Xu & Aggett, 1992).

Mineral Content

The mineral content (mg/100g) of iron, zinc, and calcium were analyzed in five commercially available Indian baby foods. The results of the analyses are shown in Table A.

The concentration of iron was highest (17.6 mg/100g) in Nestrum Rice and lowest (8.0 mg/100g) in wheat-based products Cerelac Wheat. Zinc content was highest in Cerelac Wheat (8.5mg/100g) and lowest in Nestrum Rice (2.8 mg/100g). The concentration of calcium was highest (333g/100mg) in Dexolac Rice and lowest (97.5mg/100g) in Nestrum Rice.

Studies indicate that commercial infant foods are produced from low extraction cereal flours to lower the phytic acid content present in the outer aleurone layer in rice and wheat. The processing (refining, milling, or polishing) of cereals removes some of the essential minerals.

All of the baby foods used in the present study have been fortified with minerals and vitamins by the manufacturers as indicated on the nutritional label of the food containers. However, the iron, zinc, and calcium content of wheat is higher than that of rice (Kumar & Kapoor, 1984; Aykroyd & Doughty, 1970; Pennington, 1994). In the present study the highest iron content found was in rice-based food, nestrum rice, due to the fact that it had been fortified with iron salts. Food legumes are considerably richer in zinc, iron, and calcium (Salunkhe & Kadam, 1989). The baby food, Farex, which is comprised of wheat and mung bean, has a higher content of zinc than the rice-based foods, Nestrum Rice and Dexolac Rice. However, it is lower than the two wheat-based products (Cerelac). This may be related to the amount of fortificant used in the products. The concentration of calcium was highest in the wheat-based product and Dexolac Rice, due to the presence of milk, because milk is considered to be a good source of calcium and has no phytate (Paul et. al., 1998). Nestrum Rice had the lowest concentration of calcium, being based on only rice and calcium carbonate as a fortificant.

The mineral content of the baby foods analyzed in the present study was compared to the mineral content of complementary foods used in developing countries (Gibson et al., 1998). The reported values of iron, zinc, and calcium were lower than the findings of the present study. The mineral contents (mg/100g) found in their study ranged from 0.3 – 1.6 for iron, 0.1 – 1.4 for zinc, and 4 – 106 for calcium. The lower values for minerals obtained in their study were due to the use of degermed maize flour, refined wheat flour, and polished rice, which removed most of the phytic acid. Minerals lost during processing were not replaced by mineral fortification of complementary foods used in developing countries.

Molar Ratios

The molar ratios of phytate/iron, phytate/zinc, and phytate/calcium are shown in Table B. The molar ratios of phytate/iron ranged from 0.006 in Nestrum Rice to 0.018 in Cerelac Wheat, phytate/zinc ranged from 0.019 in Cerelac Wheat and Cerelac Wheat-

Orange to 0.045 in Nestrum Rice, and phytate/calcium ranged from 0.0002 in Dexolac to 0.0008 in Nestrum Rice.

The low molar ratio values are obtained when the food has low phytate content and high mineral content, the high molar ratio values are obtained when the foods have a high content of phytate and low mineral content. The mineral and phytate contents of these baby foods depended on the ingredients of the baby foods and the amount of fortification done by the manufacturer in these baby foods.

Many published studies indicate that the molar ratios of 10 or less are usually associated with adequate zinc bioavailability and the ratios above 20 are associated with clinical or chemical evidence of zinc deficiency (Davies & Olpin, 1979). Based on the studies of Davies and Olpin (1979), deficiency of zinc will occur when more than 10 moles of phytate are present in the food per one mole of zinc. In the present study the molar ratio of phytate/zinc in all of the baby foods was less than 1. The highest value obtained was 0.045 in baby food Nestrum Rice for the phytate/zinc molar ratio indicating that 9.955 additional moles of phytate for every one mole of zinc were required to adversely effect the bioavailability of zinc. The phytate present in the baby food Nestrum Rice was not sufficient enough to complex with zinc, therefore zinc was abundantly found in terms of phytate in all of the five commercially prepared baby foods used in this study.

The phytate/zinc molar ratio in Indian baby foods was much lower than the phytate/zinc molar ratio in cooked infant Gambian foods (Paul et al., 1998) and other East African children diets (Ferguson et al., 1989). The reported molar ratios of phytate/zinc in infant Gambian foods ranged from 13-29; the ratios were greater than 10. Such levels have been considered to be detrimental to zinc bioavailability (Davies, 1982; Ellis et al., 1987; Ferguson et al., 1989; Harland, 1989). The low phytate/zinc molar ratios of the present study were less than 1.0, therefore they were below the level of concern. Gibson et al. (1991) reported similar findings, where the phytate/zinc ratios were below the level of concern. When comparing the phytate/zinc molar ratios of dietary components between different studies, some variability can be expected due to different analytical methods used for measuring phytates in foods, and the effects of processing and cooking.

According to Morris and Ellis (1985) the phytate/calcium molar ratio of 0.2 and above, may indicate a risk of calcium deficiency. Based on the study of Morris and Ellis, deficiency of calcium will occur when 0.2 or more moles of phytate were present in the baby food. In the present study the molar ratio of phytate/calcium was less than 0.2. The highest value obtained was 0.0008 in baby food Nestrum Rice for phytate/calcium molar ratio indicating that 0.1992 additional moles of phytate for every one mole of calcium were required to adversely effect the bioavailability of calcium. The phytate present in the baby food Nestrum Rice was not sufficient enough to complex with calcium. Therefore calcium was abundantly found in terms of phytate in all of these commercially prepared Indian baby foods used in the study.

Due to the limited number of studies reported in the literature on molar ratios on the phytate/iron, bioavailability of iron in Indian baby foods cannot be predicted. In comparing the molar ratios of phytate/iron in the present study to the phytate/iron molar ratios of complementary foods for infants in developing countries (Gibson et. al., 1998), the reported values were much higher (0.3-28.5mg/100g) than the phytate/iron molar ratios in Indian baby foods.

Proximate Composition

The proximate analyses (% moisture, protein, fat, and ash) of commercially prepared Indian baby foods are presented in Table A. The protein content (g/100g) ranged from 7.58 in Nestrum Rice to 16.8 in Farex (Wheat & Mung Bean). The protein contents of all of the baby foods in this study were higher (6 g/100g) than that of human milk. The protein content of wheat-based baby foods were higher than those of rice-based baby foods due to the fact that wheat contains considerably more protein than rice. Though Dexolac Rice being a rice-based baby food, it still had a higher protein content due to the presence of milk solids. The protein content of rice is 7.5 g/100g and wheat ranges from 10.5 g/100g – 13.8 g/100g (Aykroyd & Doughty, 1970).

Fat content (g/100g) of Indian baby foods ranged from 0.69 in Nestrum Rice to 8.8 in Farex (Wheat & Mung Bean). The fat concentration was higher in wheat-based baby foods than in rice-based baby foods. However, among all of the baby foods, Nestrum Rice had the lowest fat concentration, as rice was the only ingredient to provide fat, whereas in Dexolac in addition to rice, milk solids also contributed to fat content.

The ash (g/100g) content of Indian baby foods ranged from 0.57 in Nestrum Rice to 7.4 in Dexolac Rice. The moisture content (g/100g) in the Indian baby foods ranged from 3.7 in Farex to 10.89 in Cerelac Wheat. The total carbohydrates (g/100g) content in Indian baby foods ranged from 61.4 in Cerelac Wheat Orange to 84.1 in Nestrum Rice.

The nutritional composition of minerals and proximate contents in commercially prepared Indian baby foods stated on the food containers used in this study are shown in Table C. When the analyzed values of mineral content and proximate analysis in commercially prepared Indian baby foods used in the study were compared with their corresponding levels of minerals content and proximate analysis stated on the baby food containers, calcium had significantly different values. This could be due to the use of different procedures in analyzing the calcium content in the baby foods or due to improper proportions used by the manufacturer. Similar observations have also been made by Dodd and Ratani (1991) and Gillies & Neil (1985).

Conclusion

The levels of phytate, iron, zinc, and calcium content in five commercially available Indian baby foods varied. The phytate/zinc and phytate/calcium molar ratios were well below the critical ratios for both zinc and calcium. The phytate content of the five commercially available Indian baby foods was too low to inhibit these essential minerals and make them unavailable. Although, the molar ratio of phytate/iron has not been established, the iron content in all of the Indian baby foods appeared to be adequate for the daily allowances recommended for infants over the age of 6 months.

Table A. Phytate, Iron, Calcium, Zinc, and Proximate Composition in Five Commercially Prepared Indian Baby Foods.*

	Baby Food				
Composition	Nestrum	Cerelac[b]	Cerelac[c]	Farex	Dexolac
Moisture (g)	7.03	10.89	10.72	3.7	4.48
Protein (g)	7.58	15	15.46	16.8	15.1
Fat (g)	0.69	8.3	8.5	8.8	8.5
Ash (g)	0.57	2.6	3.9	4.2	7.4
Total CHO[a] (g)	84.1	63.2	61.4	66.5	66.7
Phytate (mg)	1.30	1.62	1.61	1.97	1.22
Calcium (mg)	97.5	312.5	287.2	331.1	333.0
Iron (mg)	17.6	8.0	8.33	16.04	11.2
Zinc (mg)	2.8	8.5	8.2	6.27	4.5

* per 100gm
[a] Calculated by difference
[a] CHO = Carbohydrate
[b] Cerelac (Wheat)
[c] Cerelac (Wheat/Orange)

Table B. Molar Ratios for Indian Baby Foods.

	Baby Food				
Molar Ratios[a]	Nestrum	Cerelac[b]	Cerelac[c]	Farex	Dexolac
Phytate/Iron (x 10^{-3})	6.0	18.0	17.0	11.0	9.0
Phytate/Zinc Iron (x 10^{-3})	45.0	19.0	19.0	31.0	21.0
Phytate/Calcium Iron (x 10^{-3})	0.8	0.3	0.3	0.4	0.2

[a] Moles of phytate/Moles of Mineral
[b] Cerelac (Wheat)
[c] Cerelac (Wheat/Orange)

Table C. Nutrient values of Five Commercially Prepared Indian Baby Foods from the Nutrition Label.*

	Baby Food				
Composition	Nestrum	Cerelac[b]	Cerelac[c]	Farex	Dexolac
Protein (g)	6	15.5	15.5	15.5	13.5
Fat (g)	0.6	9	9	9	7.5
Ash (g)	0.7	2.7	3.2	3.5	-
Total CHO[a] (g)	88.7	68.7	67.4	66.8	72.5
Calcium (mg)	120	570	460	750	750
Iron (mg)	18.5	7.5	7.5	15	10
Zinc (mg)	-	-	-	4.5	3.5

* per 100gm
[a] CHO = Carbohydrate
[b] Cerelac (Wheat)
[c] Cerelac (Wheat/Orange)
\- (Not present on the label)

Table D. Ingredients of Indian Baby Foods.

Product		Ingredients
Name	Type	
Nestrum (Rice)		Rice Flour, Sucrose, Calcium Carbonate, Iron Salt, Vitamins
Cerelac (Wheat)	Milk Cereal	Wheat Flour, Sucrose, Partially Skimmed milk, edible vegetable oils, Vitamins, minerals
Cerelac (Wheat & Orange)	Milk Cereal	Wheat Flour, Orange concentrate, Sucrose, Partially Skimmed milk, edible vegetable oils, Vitamins, minerals
Farex (Wheat & Mung Bean)	Milk Cereal	Wheat Flour, Mung bean, Sugar, Milk Solids, Vitamins, minerals
Dexolac (Rice)	Milk Cereal	Extruded Rice Flour, Partially Skimmed milk solids, Sucrose, Vitamins, minerals

References

Ashturkar, P.B., Pande, V., & Reddy S. (1992). Development and Evaluation of weaning food formulations. *Journal of Food Science & Technology*, **29**, No 3, 197-198.

Association of Official Analytical Chemists (1990). *Official methods of Analysis of AOAC International* (15th ed.), Arlington, VA: Authors.

Association of Official Analytical Chemists (1995). *Official methods of Analysis of AOAC International* (16th ed.), Gathersburg, MD: Authors.

Aykroyd, T., & Doughty, S. (1970). *Nutritional Value of Cereals*, St. Paul: American Association of Cereal Chemists, Inc.

Davies, N. T. (1982). Effects of phytic acid on mineral availability. In: *Dietary Fibre in Health and Disease* (*edited* by G. V. Vahony, & O. Kritchevsky). Pp 105-116, New York: Plenum.

Davies, N. T., & Olpin, S. E. (1979). Studies on the phytate: zinc molar content in diets as a determinant of zinc availability to young rats. *British Journal of Nutrition*, **41**, 591-603.

Desikachar, H. S. R. (1980). Development of weaning foods with high calorie density and low paste viscosity using traditional technologies. *Food Nutrition Bulletin*, **2**, 21.

Dodd, N. S., & Datta, S. (1988). Effect of the socio-economic level on the infant weaning practices in Maharashtrians. *The Indian Journal of Nutrition Dietetics*, **25**, 259-623.

Dodd, N. S., & Ratani, J. (1991). Mineral content of selected commercial infant and weaning foods. *Indian Journal of Dairy Science*, **44**, 122-126.

Ellis, R., Kelsay, J.L, Reynolds, R.D., Morris, E. R., Moser, P. B., & Frazier, C. W. (1987). Phytate: zinc and phytate x Calcium:zinc millimolar ratios in self-selected diets of Americans, Asian Indians, and Nepalese. *Journal of The American Dietetic Association*, **87** (8), 1043-1047.

Ferguson, E. L., Gibson, R. S., Thompson, L. U., & Ounpuu, S. (1989). Dietary calcium, phytate, and zinc intakes and the calcium, phytate, and zinc molar ratios of the diets of a selected group of East African children. *American Journal of Clinical Nutrition*, **50**, 1450-1456.

Fox, E. T., Eagles, J., & Fair weather – Tait, S. J. (1998). Bioavailability of iron glycine as a fortification in infant foods. *American Journal of Clinical Nutrition*, **67**, 664-668.

Gibson, R. S., Ferguson, E. L., & Lehrfeld, J. (1998). Complementary foods for infant feeding in developing countries their nutrient adequacy and improvement. *European Journal of Clinical Nutrition*, **52**, 764-770.

Gibson, R. S., Smit-Vanderkooy P. D., & Thompson, L. (1991). Dietary phytate calcium/zinc millimolar ratios and zinc nutriture in some Ontario preschool children. *Biological Trace Elements Research*, **30**, 87-94.

Gillies, M. E., & Neil, A. E. (1985). Minerals in infants foods. *New Zealand Medical Journal*, **98**, 788-868.

Gustafson, E-L., & Sandberg, A-S. (1995). Phytate reduction in brown beans. *Journal of Food Science*, **60**, 149-152.

Hallberg, L., Brune, M., & Rossander, L. (1989). Iron absorption in man: ascorbic acid and dose dependent inhibition by phytate. *American Journal of Clinical Nutrition*, **49**, 140-144.

Harland, B. F. (1989). Dietary fibre and mineral availability. *Nutrition Research*, **Rev 2**, 133-147.

Harland, B. F., & Morris, E. R. (1995). Phytate: A good or a bad component. *Nutrition Research*, **15**, 733-755.

Harland, B. F., & Oberleas, D. (1987). Phytate in Foods. *World Review in Nutrition and Dietetics*, **52**, 235-259.

Jones, W. M. (1949). *Inorganic chemistry*, Pp. 377. Philadelphia: Blakiston.

Kumar, V., & Kapoor, A. C. (1984). Trace mineral composition of different varieties of cereals and legumes. *The Indian Journal of Nutrition and Dietetics*, **21**, 137.

Kumar, K. G., Venkataraman, L. V., Jaya, T. V., & Krishnamurthy, K. S. (1978). Cooking characteristics of some germinated legumes: changes in phytin, ca++, and pectins. *Journal of Food Science*, **43**, 85.

Morris, E. R., & Ellis, R. (1985). Bioavailability of dietary calcium: effect of phytate on adult men consuming non-vegetarian diets; in Nutritional bioavailability of calcium. *Journal of American Chemists Society*, **275**, 63-72.

Paul, A. A., Bates, J. B., Prentice, A., Day, C. D., & Tsuchiya, H. (1998). Zinc and phytate intake of rural Gambian infants: contributions from breast milk and weaning foods. *International Journal of Food Science and Nutrition*, **49**, 141-155.

Pennington, J. A. T. (1994). Bowes and church's food values of portion commonly used (edited by J.B. Lippincott Co.). Pp 153-169. New York:Perennial Books.

Salunkhe, D. K., & Kadam, S. S. (1989). *Handbook of world food legumes: Nutritional Chemistry, processing, technology, and utilization*, Vol. **1**, Boca Raton, FL:CRC Press.

Sandberg, A. S., Carlsson, N. G. & Svanberg, W. (1988). In Vitro studies of inositol tri-tetra- penta, and hexaphosphate as potential iron absorption inhibitors. *Bioavailability Abstracts*, UK.

Sehgal, S., Kapoor, A. C., & Taneja, A. (1989). Prevalence of food fads and fallacies regarding infant feeding among laborer and urban mothers. *The Indian Journal of Nutrition Dietetics*, **26**, 206-210.

Whitney, N. E., & Rolfes S. R. (1993). The trace minerals. In *Understanding Nutrition.* Pp. 442. St. Paul: West Publishing Co.

Xu, P. P. J., & Aggett P. J. (1992). Recent advances in methodology for analysis of phytate and inositol phosphates in foods. *Progress in Food Nutrition and Science*, **16**, 245-262.

CHAPTER 3

SUMMARY AND RECOMMENDATIONS

Summary

Cereal and legume-based products are commonly available in India. They form the bulk of the infant's diet. These plant foods are the main source of minerals and other nutrients. The presence of a naturally occurring antinutritional substance called phytate occurs in plant food, with the highest level in legumes and cereal grains, and it is well documented that phytate interferes with mineral utilization. Since calcium, zinc, and iron are important nutrients for an infant's growth, these minerals in food should be in a highly bioavailable form. However, there is not much information available on the levels of phytate, and essential minerals like calcium, iron, zinc and their molar ratios for bioavailability of minerals in Indian baby foods.

The present study was initiated to determine the phytate and mineral content (iron, zinc, and calcium) of five commercially prepared Indian baby foods. Molar ratios of phytate/iron, phytate/zinc, and phytate/calcium were calculated to determine the bioavailability of minerals in Indian baby foods.

The results of this study indicated that the amount of phytate present in the baby foods was low. Mineral content of the baby foods appeared to be adequate for daily allowances recommended for infants over the ages of six months. The level of phytate and the minerals iron, calcium, and zinc in Indian baby foods varied depending on the ingredients and type of mineral fortification performed during processing by the manufacturer.

Based on the established and published molar ratios, 10 or less for phytate/zinc and 0.2 or less for phytate/calcium, this study concluded that in these five commercially available Indian baby foods, zinc and calcium were relatively bioavailable. The phytate/iron molar ratios were also relatively low and the fact that the iron content in all the five Indian baby foods appeared to be adequate for the daily recommendation for infants over the age of 6 months. As phytate concentration was low in all of the baby foods analyzed, it was concluded that iron was also bioavailable in these baby foods.

Recommendations

Although determination of molar ratios based on chemical analysis of phytate, calcium, zinc and iron is a relatively inexpensive method of predicting bioavailability of essential minerals, the results are only estimates. The results of molar ratios of phytate/minerals in commercially available Indian baby foods analyzed in this study determined the bioavailability of metabolic minerals, but this does not assure that the infants who are consuming this food will absorb the minerals. Using human subjects is time consuming and expensive, but these studies are more reliable for predicting the bioavailability of minerals in baby foods. Therefore, further analysis needs to be performed to determine the effect of phytate on bioavailability of minerals in Indian baby foods based on research utilizing human subjects.

It is important to recognize that this study has only considered the bioavailability of zinc and calcium in the baby foods in terms of only phytate, there are other antinutritional factors like fiber, polyphenols, and oxalates, which inhibit minerals and play a major role in mineral bioavailability. Therefore, further work needs to be done with respect to other anti-nutritional factors present in the commercially prepared Indian baby foods, which effect the bioavailability of zinc, calcium, and iron.

Besides the role of phytate in the inhibition of mineral absorption in humans, mineral-mineral interactions in foods also determine the bioavailability of minerals. Foods high in calcium, potentiates the complexation of zinc with phytate. Some studies suggest that the phytate x calcium/zinc molar ratio is a better indicator of zinc utilization, Therefore in the future, predicting zinc bioavailability from high calcium foods, molar ratios of phytate x calcium/zinc could be pursued.

According to Morris and Ellis (1985) the phytate/calcium molar ratio of 0.2 and above may indicate the risk of calcium deficiency. There are not enough studies available supporting the fact that the value of phytate/calcium molar ratio of 0.2 or less is an indicator of calcium adequacy, therefore further research needs to be performed in this area. Also, further investigation is needed on the molar ratio of phytate/iron, in order to estimate the bioavailability of iron in foods.

References

Abernathy, R. H., Paulson, G. M., & Ellis, R. Jr. (1973). Relationship among phytic acid, phosphorus, and zinc during maturation of winter wheat. Journal of Agricultural Food Chemistry, 21, 282.

Al-Othman, A.M., Khan, M. A., & Al-Kanhan, M. A. (1997). Nutritional evaluation of some commercial baby foods consumed in Saudi Arabia. International Journal of Food Science and Nutrition, 48, 229-236.

Adsule, R. N., Lawande, K. M., & Kadam, S. S. (1984). Malted Sorghum: a potential source for low cost weaning food. In D. K. Salunkhe, J. K. Chavan, & S. J. Jadhav (Eds.), Nutritional and processing quality of Sorghum (Pp 187). New Delhi: Oxford & IBH.

Aggett, P. J., Atherton, D.J., More, J., Davey, J., Delves, H. T., & Havies, H. T. (1980). Symptomatic zinc deficiency in a breast fed preterm infants. Archives of Disable Children, 55, 547-550.

Anderson, R. J. (1914). Journal Biological Chemistry, 17, 141.

Asada, K., Tanaka, K., & Kasai (1978). Formation of phytic acid in cereal grain. Annalytical Biochemistry, 165(2), 4.

Ashturkar, P.B., Pande, V., & Reddy, S. (1992). Development and Evaluation of weaning food formulations. Journal of Food Science & Technology, 29(3), 197-198.

Association of Official Analytical Chemists (1990). Official methods of Analysis of AOAC International (15th ed.), Arlington, VA: Author.

Association of Official Analytical Chemists (1995). Official methods of Analysis of AOAC International (16th ed.), Gathersburg, MD: Author.

Aykroyd, T., & Doughty, S. (1970). Nutritional Value of Cereals. St. Paul: American Association of Cereal Chemists, Inc.

Betschart, A. A. (1988). Nutritional Quality of wheat and wheat foods. St. Paul: American Association of Cereal Chemists Inc., 91-95.

Blom, I., Jameson, S., Krook, F., Larsson-stymne, B., & Waranne, L. (1980). Zinc deficiency with transitory acrodermaticts enteropathica in a boy of low birth weight. British Journal of Dermatology, 104, 459-464.

Booth, R. G., Carter, R. H., Jones, C. R., & Moran, T. (1941). The Nations food II: Cereal as food chemistry of wheat and wheat products. Journal of Soc. Chem. Ind. (London), 60, 903-908.

Bressani, R., Elias, L. G., & Altschul, M. (Eds.) (1974). Legume foods in new protein foods, Vol. 1, New York: Academic Press, 230.

Casey, C. E., Walravens, P. A., & Hambidge, K. M. (1981). Availability of zinc: loading tests with human milk, cow's milk and infant formula. Pediatrics, 68, 394-396.

Chatterjee, S. R., & Abrol, Y.P. (1975). Amino acid composition of new varieties of cereals and pulses and nutritional potentials of cerela-pulse combination. Journal of Food Science and Technology, 12, 221-224.

Chen, L. H., & Pan, S. H. (1977). Decrease of phytates during germination of pea seeds (pisum Sativum). Nutrition Report Int., 16, 125.

Clydesdale, M., F. (1988). Minerals: Their chemistry and fate in food. New York: Marcel Dekker, Inc., 68-69.

Cook, J. D., Reddy, M. B., Burri, J., Juillerrat, M. A., & Hunell, R. F. (1997). The influence of different cereal grains on iron absorption from infant cereal foods. American Journal of Clinical Nutrition, 65, 964-969.

Cosgrove, D. J. (1963). Chemistry of phytate. Journal of Biochemistry, 89, 172.

Cosgrove, D. J. (1966). The chemistry and biochemistry of inositol polyphosphates. Reviews in pure applied chemistry, 16, 209.

Davidsson L., Galan P., & Cherouvrier F. (1997). Bioavailability in infants of iron from infant cereals: effect of dephytinization. American Journal of Clinical Nutrition, 65, 916-920.

Davies, N. T. (1982). Effects of phytic acid on mineral availability. In: *Dietary Fibre in Health and Disease* (*edited* by G. V. Vahony, & O. Kritchevsky). Pp 105-116, New York: Plenum.

Davies, N. T., & Olpin, S. E. (1979). Studies on the phytate: zinc molar contents in diets as a determinant of zinc availability to young rats. British Journal of Nutrition, 41, 591-603.

deBoland, A. R., Garner, G. B., & O'Dell, B. L. (1975). Identification and properties of phytate in cereal grains and oilseed products. Journal of Agriculture and Food Chemistry, 23, 1186.

Desikachar, H. S. R. (1980). Development of weaning foods with high calorie density and low paste viscosity using traditional technologies. Food Nutrition Bulletin, 2, 21.

Dodd, N. S., & Datta, S. (1988). Effect of the socio-economic level on the infant weaning practices in Maharashtrians. The Indian Journal of Nutrition Dietetics, 25, 259-623.

Dodd, N. S., & Ratani, J. (1991). Mineral content of selected commercial infant and weaning foods. Indian Journal of Dairy Science, 44, 122-126.

Ellis, R., Kelsay, J. L., Reynolds, R.D., Morris, E. R., Moser, P. B., & Frazier, C. W. (1987). Phytate: zinc and phytate x Calcium:zinc millimolar ratios in self-selected diets of Americans, Asian Indians, and Nepalese. Journal of The American Dietetic Association, 87 (8), 1043-1047.

Erdman, J. W., Jr. (1979). Oilseed phytates: Nutritional implications. Journal of the American Oil Chemists' Society, 56, 736-741.

Erdman, J. W., & Forbes, R. M. (1977). Mineral bioavailability from phytate – containing foods. Food Product Development, 11(10), 46.

Erdman, J. W., & Thompson, D. B. (1982). Structural model for phytate: Implications for phytic acid analysis. Cereal Chemistry, 59, 525-528.

Fairweather-Tait, S. J., & Caprez, A. (1982). A comparison between the effect of added sodium phytate and endogenous phytate in bran and vats on zinc absorption. Proceedings in Nutrition Society, 41, 132.

Fennema, O. R. (1996). Minerals. In Food chemistry (2nd ed.). New York: Marcel Dekker.

Ferguson, E. L., Gibson, R. S., Thompson, L. U., & Ounpuu, S. (1989). Dietary calcium, phytate, and zinc intakes and the calcium, phytate, and zinc molar ratios of the diets of a selected group of East African children. American Journal of Clinical Nutrition, 50, 1450-1456.

Food and Agriculture Organization of the United Nations. (1992). Nutrition and Development a global assessment, Rome.

Food and Agriculture Organization of the United Nations. (1984). Production Yearbook, Rome.

Fox, E. T., Eagles, J., & Fair weather – Tait, S. J. (1998). Bioavailability of iron glycine as a fortification in infant foods. American Journal of Clinical Nutrtition, 67, 664-668.

Gibson R. S. (1994). Zinc Nutrition in developing countries. Nutrition Research Reviews, 7, 151-173.

Gibson, R. S., Ferguson, E. L., & Lehrfield, J. (1998). Complementary foods for infant feeding in developing countries their nutrient adequacy and improvement. European Journal of Clinical Nutrition, 52, 764-770.

Gibson, R. S., Smit-Vanderkooy P. D., & Thompson, L. (1991). Dietary phytate calcium/zinc millimolar ratios and zinc nutriture in some Ontario preschool children. Biological Trace Elements Research, 30, 87-94.

Gillies, M. E., & Neil, A. E. (1985). Minerals in infants foods, New Zealand Medical Journal, 98, 788-868.

Gillooly, M., Bothwell, T. H., & Tonance, J. D. (1984). The Effects of organic acids, Phytates and polyphenols on the absorption of iron form vegetables. British Journal of Nutrition, 49, 331-342.

Graf, E. & Eaton, J. W. (1993). Suppression of colonic cancer by dietary phytic acid. Nutrition Cancer, 19, 11-20.

Gustafson, E-L., & Sandberg, A-S. (1995). Phytate reduction in brown beans. Journal of Food Science, 60, 149-152.

Hallberg, L., Brune, M., & Rossander, L. (1989). Iron absorption in man: ascorbic acid and dose dependent inhibition by phytate. American Journal of Clinical Nutrition, 49, 140-144.

Hallberg, L., & Rossander, L. (1982). Effect of soy protein on nonheme iron absorption in man. American Journal of Clinical Nutrition, 36, 514-520.

Hallberg, L., Rossander, L., & Kanberg, A. B. (1987). Phytates and the inhibitory effect of bran on iron absorption in man. American Journal of Clinical Nutrition, 45, 988-996.

Hambidge, K. M. (1988). Mild zinc deficiency in human subjects. In C. F. Mills (Eds), Zinc in human biology (pp. 160-170). London: Springer-Verlag.

Hambidge, K. M., & Silverman, A. (1973). Pica with rapid improvement after dietary zinc supplementation. Archives of Disable Child, 48, 67-568.

Hambidge, K. M., Walravens, P. A., Casey, C., Brown, R. M., & Bender, C. (1979). Plasma zinc concentrations of breast fed infants. Journal of Pediatrics, 94, 607-608.

Harland, B. F. (1978). Interactions of fiber and other nutrients. Medical Challenge, 10, 30-44.

Harland, B. F. (1989). Dietary fibre and mineral availability. Nutrition Research, Rev 2, 133-147.

Harland, B. F., & Morris, E. R. (1995). Phytate: A good or a bad component. Nutrition Research, 15, 733-755.

Harland, B. F., & Oberleas, D. (1985). Phytate and zinc contents of coffees, cocoas, and teas. Journal of Food Science, 50, 832-842.

Harland, B. F., & Oberleas, D. (1987). Phytate in Foods. World Review in Nutrition and Dietetics, 52, 235-259.

Hazell, T., & Johnson, I. T. (1987). In vitro estimation of iron availability from a range of plant foods: Influence of phytate, ascorbate and citrate. British Journal of Nutrition, 57, 223-233.

Heaney, R. P., & Weaver, C. M. (1989). Oxalate: Effect on calcium absorbability. American Journal of Clinical Nutrition, 50, 830-832.

Hemminiki, E. (1995). Impact of Iron fortification of milk formulas on infants growth and health. Nutrition Research, 15(4), 491-503.

Herbert, V. (1987). Recommended dietary intakes (RDI) of iron in humans. American Journal of Clinical Nutrition, 45, 679-686.

Holub, B. J. (1992). The nutritional importance of inositol and the phospho inositides. New England Journal of Medicine, 326, 1285-1286.

Jones, W. M. (1949). Inorganic chemistry. Philadelphia: Blakiston. 377.

Jorgensen, E., Anderson, O., & Nielsen, G. (1946). The effect of phytic acid on the absorption of calcium and phosphorous. Biochemistry Journal, 40, 555.

Kelly, J. F. (1985). Stabilization of rice polish. U.S. Patent 4, 158, 066.

Kennedy, B. M., & Schelstraete, M. (1975). Phytate in cereals. Cereal Chemistry, 52, 173.

Kent-Jones, D. W., & Amos, A. J. (1957). Modern Cereal Chemistry. Liverpool: Northern Publishing. 492.

Khan, M. A., & Kissana, A. S. (1985). Nutritional evaluation of some baby foods in Pakistan. Journal of Food Science and Agriculture, 36, 1271-1274.

Kumar, K. G., Venkataraman, L. V., Jaya, T. V., & Krishnamurthy, K. S. (1978). Cooking characteristics of some germinated legumes: changes in phytin, ca++, and pectins. Journal of Food Science, 43, 85.

Kumar, V., & Kapoor, A. C. (1984). Trace mineral composition of different varieties of cereals and legumes. The Indian Journal of Nutrition and Dietetics, 21, 137.

Kuramoto, Y., Igarashi, Y., & Tagami, H. (1991). Acquired zinc deficiency in breast fed infants. Seminar in Dermatology, 10, 309-312.

Lam, N. T., Gross, R., Sastramidjojo, S. & Schultink, W. (1997). Relative bioavaialbility in adults of iron from a soybean product intended for complementary feeding of infants. International Journal of Food Science and Nutrition, 48, 359-364.

Linder, C. M. (1991). Nutritional Biochemistry and metabolism with clinical applications (2nd ed.). New York: Elsevier Science.

Likuski, H. J. A., & Forbes, R. M. (1965). Mineral utilization in the rat: Effect of calcium and phytic acid on the utilization of dietary zinc. Journal of Nutrition, 85, 230-234.

Lolas, G. M., & Markakis, P. (1975). Journal of Agriculture and Food Chemistry, 23, 13.

Lolas, G. M., Palamidas, N., & Markakis, P. (1976). Cereal Chemistry, 53, 867.

Loomis, W. F. (1970). Rickets. Scientific American, 223, 77-91.

Lonnerdal, B. O., Jayawickrama, L., & Lien, E. L. (1999). Effect of reducing the phytate content and of partially hydrolyzing the protein in soy formula on zinc and copper absorption and status in infants rehus monkeys and rat pups. American Journal of Clinical Nutrition 69(3), 490-496.

Lonnerdal, B., Stanislowski, A. G., & Hurley, L. S. (1980). Isolation of low molecular weight zinc-binding ligand from milk. Journal of inorganic biochemistry,12, 71-78.

Lott, J. N. A. & Buttrose, M. K. (1978). Globoids in protein bodies of legume seed cotyledons. Australian Journal of Plant Physiology, 5, 89.

Lynch, S. R., Beard, J. L., & Dassenko, S. A. (1984). Iron absorption from legumes in human. American Journal of Clinical Nutrition, 40, 42-47.

Lynch, S. R., Dassenko, S. A., Morck, T. A., Beard, J. L., & Cook, J. D. (1985). Iron availability from infant food supplements. American Journal of Clinical Nutrition, 41, 13-20.

Maddaiah, V. T., Kurnick, A. A., & Reid, B. L. (1964). Phytic acid studies. Proceedings of Society for Experimental Biology & Medicine, 115, 391-393.

Maga, A. Joseph (1982). Phytate: Its chemistry, occurrence, food interactions, nutritional significance, and method of analysis. Journal of Agriculture and Food Chemistry, 30(1), 1-9.

Margen, S. (1992). The Wellness Encyclopedia of Food and Nutrition (2nd ed.). New York:Rebus.

Morck, T. A., Lynch, S. R., Skikne, B. S., & Cook, J. D. (1981). Iron avaialbility from infant food supplements. American Journal of Clinical Nutrition, 34, 2630-2634.

Morris, E. R., & Ellis R. (1985). Bioavailability of dietary calcium: effect of phytate on adult men consuming non vegetarian diets. In nutritional bioavailability of calcium. Journal of American Chemists Society,275, 63-72.

Moss, H. J. (1973). Quality standards for wheat varieties. Journal of Australian Institute of Agriculture Science, 39, 109-115.

Nahapetian, A. & Bassiri, A. (1976). Variation in concentrations and interrelationships of phytate phosphorus, magnesium, calcium, zinc, and iron in wheat varieties during two years. Journal of Agricultural Food chemistry, 24, 947.

Naik, M. S., & Narayana, N. (1959). Indian Journal of Applied Chemistry, 22, 239.

Nolan, K. B., Duffin, P. A., & McWeeny, D. J. (1987). Effect of phytate on mineral bioavailability. In Vitro Studies on Mg^{2+}, Ca^{2+}, Fe^{3+}, Cu^{2+}, and Zn^{2+} (also Cd^{2+}) Solubilities in presence of phytate. Journal of Science and Food Agriculture, 40, 79-85.

Oberleas, D. (1973). Toxicants Occurring naturally in Foods. Washington, D.C.: National Academy of Sciences, 363.

Oberleas, D., (1975). Factors influencing availability of minerals. Proceedings for Western Hemisphere Nutrition Congress. Acton: Publishing Science Group, 156-161.

Oberleas, D. (1983a). The role of phytate in zinc bioavailability and homeostasis: in nutritional bioavailability of zinc. Journal of American Chemists Society, 145-158.

Oberleas, D. (1983b). Phytate content in cereals and legumes and methods of determination. Cereal Foods World, 28, 352-357.

Oberleas, D., & Harland, B. F. (1981). Phytate content of foods: Effect on dietary zinc bioavailability. Journal of American Dietetic Association, 79, 433-436.

Oberleas, D., & Prasad, A. S. (1976). Trace Elements in Human Health and disease. New York: Academic Press, 155-162.

O'Dell, B. L. (1979). Soy Protein and Human Nutrition. New York: Academic Press.

O'Dell, B. L. (1969). Effect if dietary components upon zinc availability. American Journal of Clinical Nutrition, 22, 1315-1322.

O'Dell, B. L., & deBoland, A. R. (1976). Complexation of phytate with proteins abd cation in corn germ and oilseed meals. Journal of Agriculture and Food Chemistry, 24, 804-807.

Orth, R. A., & Schellenberger J. A. (1988). Origin, production, and utilization of wheat. St. Paul:American Association of Cereal Chemists Inc, 1-13.

Paul, A. A., Bates, J. B., Prentice, A., Day, C. D., & Tsuchiya, H. (1998). Zinc and phytate intake of rural Gambian infants: contributions from breast milk and weaning foods. International Journal of Food Science and Nutrition, 49, 141-155.

Pawar, V. D., & Ingle, V. M. (1987). Production of quick-cooking moth beans (phaseolus A conitifolius JACQ) II Phytate Phosphorous and minerals. The Indian Journal of Nutrition Dietetics, 24, 142.

Pennington, J. A. T. (1994). Bowes and church's food values of portion commonly used (edited by J.B. Lippincott Co.), Pp 153-169. New York:Perennial Books.

Pfeffer, E. (1872). Jahrb. Wiss. Bot., 8, 429.

Pipes, P. L. (1977). Nutrition in Infancy and Childhood. St. Louis: C. V. Mosley Co., 63-65.

Pomeranz, Y. (1988). Wheat Chemistry and Technology, Vol. 1.St, Paul: American Association of Cereal Chemists Inc., 12.

Pretlow, T. P., O'Riordan, M. A., Somich, G. A., Amini, S. B., & Pretlow, T. G. (1992). Aberrant crypts correlate with tumor incidence in F 344 rats treated with azomymethane and phytate. Carcinogen, 13, 1509-1512.

Purvis, G. A., & Bartholmey, S. J. (1988). Infant feeding practices: Commercially prepared baby foods. Philadelphia:The CV Mosley Co., 399-407.

Rackis, J. J., & Anderson, R. L. (1977). Mineral bioavailability in soy protein products. Food Product Development, 11, 38.

Rajalakshmi, R. (1976). Preschool Child Nutrition: Patterns, prevalence, Prevention. Baroda Journal of Nutrition, 3, 1-129.

Reddy, N. R., & Salunkhe, D. K. (1981). Journal of Food Science, 46, 564.

Reddy, N. R., Sathe, S. K., & Salunkhe, D. K. (1982). Phytates in legumes and cereals. Advance Food Research, 28, 1.

Reinhold, J. G, Parsa, M., Karimian, N., Hammick, J. W., & Ismail-Beigi, F. (1975). Availability of zinc in unleavened and leavened whole meal breads as measured by solubility and uptake by rat by intestine in vitro. Journal of Nutrition, 104, 976-982.

Roxas, B. V., Intengann, C. L., & Juliano, B. O. (1976). Protein content of milled rice and nitrogen retention of preschool children fed rice-mung bean diet. Nutrition Report International, 14, 203.

Salunkhe, D. K. (1982). Legumes in human nutrition: current status and future research needs. Current Science, 51, 387.

Salunkhe, D. K., Bolin, H. R., & Reddy, N. R. (1991). Storage, Processing, & Nutritional Quality of fruits and vegetables, Vol. 1. Boca Raton, FL: American Association of Cereal Chemists Inc., 24.

Salunkhe, D. K., & Kadam, S. S. (1989a). Handbook of world food legumes: Nutritional Chemistry, processing, technology, and utilization, Vol. 1. Boca Raton, FL: CRC Press.

Salunkhe, D. K., & Kadam, S. S. (1989b). Handbook of world food legumes: Nutritional Chemistry, processing, technology, and utilization, Vol. II. Boca Raton, FL: CRC Press.

Samotus, B., & Schwimmer, S. (1962). Nature (London), 194, 578.

Sandberg, A. S., Carlsson, N. G. & Svanberg, W. (1988). In Vitro studies of inositol tri- tera- penta, and hexaphosphate as potential iron absorption inhibitors, *Bioavailability Abstracts*, UK.

Sandstrom, B., Cederblad, A., & Lonnerdal, B. (1983). Zinc absorption from human milk, cow's milk, and infant formulas. American Journal of Disabled Child, 137, 726-729.

Sazawal, S., Black, R. E., & Bhan, M. K. (1996). Zinc supplementation reduces the incidence of persistence diarrhea and dysentry among low socio economic children in India. Journal of Nutrition, 126, 443-493.

Scrimshaw, N. S. (1994). The consequences of hidden hunger for individuals and societies. Food Nutrition Bulletin, 51, 3-24.

Sehgal, S., Kapoor, A. C., & Taneja, A. (1989). Prevalence of food fads and fallacies regading infant feeding among labourer and urban mothers. The Indian Journal of Nutrition Dietetics, 206-210.

Serraino, M. R., & Thompson, L. V. (1984). Removal of phytic acid and protein – phytic acid interactions in rapeseed. Journal of Agriculture and Food Chemistry, 32, 38-40.

Shaw, J. C. L. (1979). Trace elements in the fetus and young infants. American Journal of Disabled Child, 13, 1260-1258.

Simpson, K. M., Morris, E. R., & Cook, J. D. (1981). The inhibitory effect of bran on iron absorption in man. American Journal of Clinical Nutrition, 34, 1469-1478.

Singh, M., & Krikorian, A. D. (1982). Inhibition of trypsin activity in Vitro by phytate. Journal of Agriculture and Food Chemistry, 30, 799-800.

Singh, P. P., Sharma, M. C., & Sur, B. K. (1969). Value of green leaves as sources of available calcium. Indian Journal of Medical Research, 57, 204-209.

Singh, S., Ingh, H. D., & Sikka, K. C. (1968). Distribution of Nutrients in the anatomical parts of common Indian pulses. Cereal Chemistry, 45, 13.

Sobolev, A. M. (1966). On the state of phytin in the aleurone grains of mature and germinating seeds. Soviet Plant Physiology, 13, 177.

Sudarmadji, S., & Markkakis, P. (1977). The phytate and phytase of soybean tempeh. Journal of Science and Food Agriculture, 28, 381.

Tabekhia, M. M., & Luh, B. S. (1979). Effect of milling on macro- and micro-minerals and phytate of rice. Deutsche Lebensm Rundsch, 75, 57.

Tanaka, K.,Yoshida, T., & Kasai, Z. (1974). Radioautographic demonstration of the accumulation site of phytic acid in rice and wheat grains. Plant Cell Physiology, 15, 147.

Thompson, S. A., & Weber, C. W. (1981). Effects of dietary fiber sources on tissue minerals levels in chicks. Poultry Science, 60, 840-845.

Toma, R. B., & Tabekhia, M. M. (1979). Journal of Food Science, 44, 619.

Tonance, G. M., Tonance, J. D., & Bothwell, T. H. (1984). The relative effect of ascorbic acid on iron absorption form soy-based and milk based infant formulas. American Journal of Clinical Nutrition, 40, 522-527.

Verma, S. C., & Lal, B. M. (1966). Journal of Science & Food Agriculture, 17, 43.

Vohra, P., Gray, G. A., & Kratzer, F. H. (1965). Phytic acid-metal complexes. Proceedings of Society for Experimental Biology & Medicine, 120, 447.

Walker, A. R. P., Walker, B. F., & Wadvalla, M. (1968). An attempt to measure the availability of calcium in edible leaves commonly consumed by South African negroes. Ecology of Food Nutrition, 26, 65-70.

Weaver, C. M., (1998). Roles of phytate in absorption of minerals. Web based.

Whitney, N. E., & Rolfes S. R. (1993a). The trace minerals. In Understanding Nutrition. St. Paul: West Publishing Co., 422.

Whitney, N. E., & Rolfes S. R. (1993b). Recommended dietary allowance for infants. In Understanding Nutrition. St. Paul: West Publishing Co., 1.

Wilson, A. M., & Harris, G. A. (1966). Plant Physiology,41 , 1416.

Wilson, D. C., & Widdowson, E. M. (1942). Indian Res. Mem., 34.

Xu, P. P. J., & Aggett P. J. (1992). Recent advances in methodology for analysis of phytate and inositol phosphates in foods. Progress in Food Nutrition and Science, 16, 245-262.

Ziegler, E. E., & Fomon, J. S. (1996). Startegies for the prevention of iron deficiency in infant formulas and baby foods. Nutrition Reviews, 54(11), 348-354.

Zimmerman, A. W., Hambidge, M., Lepov, M. L., Greenberg, R. D., Storer, M. L., & Casey, C. E. (1982). Acrodermatitis in breast fed premature infants: evidence for a defect of mammary zinc secretion. Pediatrics, 69, 176-183.