THE EFFECTS OF UNPROCESSED AND PROCESSED OAT BRAN ON MINERAL BIOAVAILABILITY IN ADULT MEN

by

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(ABSTRACT)

The effect of comsumption of unprocessed and processed oat bran on calcium, magnesium, zinc, and copper intake, excretion and apparent retention in 19 adult males was determined using a metabolic balance study. The study was divided into preliminary, controlled feeding and follow-up periods of 4 weeks each. The controlled feeding period was subdivided into two 8 day balance periods. Subjects were randomly assigned to one of three treatments: a basal diet low in dietary fiber; basal diet supplemented with 100 g of unprocessed oat bran; or basal diet supplemented with 100 g of processed oat bran ready-to-eat (RTE) cereal.

With both oat bran treatments intakes of calcium, magnesium, and copper were increased above the current RDA, while zinc was increased, but still remained below the RDA. Urinary excretions of calcium, magnesium, and zinc were unchanged during the balance periods. Both of the bran supplemented groups were excreting significantly more fecal magnesium and zinc than the control group. Increased fecal calcium and copper excretions were seen for the processed

(RTE) group over the other two treatments. Apparent retention of calcium, zinc, and copper appear to be unaffected by oat bran supplementation. Apparent magnesium retention for the control group was significantly less than the unprocessed group during balance Period I only. During the controlled feeding period, the unprocessed group had significantly less plasma calcium and zinc than the other two treatments. Plasma magnesium was unchanged in the fiber supplemented groups during the controlled feeding period.

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INTRODUCTION

Public interest in dietary fiber has greatly increased in the past decade. It has been said that fiber deficiency is a major health concern in Westernized societies (Cummings, 1978; Vahouny, 1982; Trowell, 1978). Recent studies (NHANES II, 1976-80) and (NHIS, 1987) have estimated the mean dietary fiber intake of the United States adult population at 11.1 grams/day and 12 grams/day, respectively (Lanza et al, 1987; Block and Subar, 1992). A report prepared for the United States Food and Drug Administration recommends a fiber intake of 20-35 grams/day as part of a healthy diet (Pilch, 1987). The recent development of the Food Guide Pyramid by the United States Department of Agriculture (USDA) suggests that normal, healthy adults should consume 6 to 11 servings of breads, cereals, rice or pasta each day as the basis for following the Dietary Guidelines for Americans (USDA, 1992).

Over time, studies have indicated that dietary fiber exerts some type of diverse physio-chemical effects and appears to function in both the upper and lower gastrointestinal tract of humans (Bingham, 1987). We know that fiber alters the digestion, absorption and subsequent metabolism of various nutrients (Cummings, 1978; Vahouny,

1982; National Research Council, 1989). It has been shown that consuming increased levels of dietary fiber causes increased fecal bulk and speeds up intestinal transit time; thus having a laxative effect on the digestive system (Kelsay, 1978; Eastwood, 1978; Eastwood et al., 1986; Lockhart and Hurt, 1986). This bulking property can cause the displacement of essential nutrients away from the lumen wall. Burkitt (1971) proposed a mechanism whereby dietary fiber protected against colonic cancer by regulating the speed of colonic transit and the consistency and weight of stool, thereby diluting carcinogens and altering microbial metabolism.

Certain foods high in dietary fiber show much promise as tools in the nutritional treatment of diseases such as diabetes, hypercholesterolemia, colon carcinogenesis and diverticular disease (Cummings, 1978; Kelsay, 1978; Eastwood, 1978; Kritchevsky, 1983; Klurfeld and Kritchevsky, 1986; Sacks, 1991; Shinnick et al., 1991). The complex carbohydrates which make up dietary fiber are more slowly digested than simple sugars, making fibrous foods a good source of carbohydrate for the diabetic because of their slower digestion. Dietary fiber or a property of the fiber may act to bind bile salts and cholesterol in the intestine which alters cholesterol levels in humans (Kelsay, 1978;

Kritchevsky, 1983; Klopfenstein, 1988). Physical structure of fiber accounts in part for its hygroscopic nature, making stools soft and easily eliminated, thereby lessening the irritation to the intestinal walls in diverticular disease (Kriek et al., 1982). Proponents of dietary fiber are beginning to view it as a "nutrient" rather than as a "non-nutritional factor".

Dietary fiber may have some adverse consequences on nutritional status by interfering with the absorption of various nutrients in the diet (Klopfenstein, 1988; NRC, 1989). Dietary fiber appears to exert its most deleterious effect on the bioavailability of minerals (Van Soest, 1978b; Davies, 1979; Lockhart and Hurt, 1986). Studies have focused mainly on divalent minerals such as calcium, copper, cobalt, iron, magnesium, manganese, and zinc. Physical structure and phytates appear to bind these minerals although no mechanism has been proved. Most study results to date have concluded that there is either a decrease in absorption of minerals or that there is no change in mineral balance (Southgate, 1987; Frølich, 1993).

In light of the effects that dietary fiber has on humans, an investigation was proposed to look at the effect of feeding a fiber known for its hypocholesterolemic properties (oat bran) and studying the effect it had on

mineral bioavailability in healthy adult men. Four minerals; calcium, magnesium, zinc, and copper were studied with regard to balance and plasma levels.

REVIEW OF LITERATURE

I. Dietary Fiber

The definitions of dietary fiber (DF) has been a source of confusion for investigators for many years since the definitions may originate from a physiological, chemical or botanical point of view, or any combination of these (Spiller, 1993). Trowell, (1972) used the definition which referred to the skeletal remains of the plant cell walls in foods. This definition was further developed to focus on the unavailable polysaccharides and lignin in foods with unavailable polysaccharides being those that are not digested by the endogenous enzymes secreted into the gastrointestinal tract (Trowell et al., 1976; Southgate, 1992). This definition presents problems to analytical chemists who tend to prefer the definition proposed by Southgate (1982) which is the sum of lignin and non α-glucan (nonstarch) polysaccharides in foods. The differences in definitions have risen because fiber is not a single entity but a mixture of many complex organic substances, including resistant starch, and bioactive compounds (Lanza and Butrum, 1986; Spiller, 1991). A group of substances that share certain chemical or biological properties have traditionally been gathered under all-encompassing terms in nutrition,

thus, adding to the complexity of defined components (Spiller, 1993). The word, fiber, brings to mind a picture of long fibrils, which is in agreement with the physical characteristics of some components of DF, mainly cellulose, but not of others. The term "Plantix" has been suggested as an alternative to DF and implies that the source is a plant and that matrix formation is one of its characteristics (Spiller, 1993). Plant cell walls are the main source of DF for humans with most intake coming from cell walls in foods such as vegetables, fruits and cereal products. The principle components of plant cell walls are listed in Table 1 and are complex polysaccharides, glycoproteins and lignin (Selvendran, 1984). The list of components making up plant cell walls has increased as fractionation analyses have evolved. Recently, some researchers have started to differentiate fractions of dietary fiber by name, such as nonstarch polysaccharides (NSP), resistant starch (RS), and lignin. Nonstarch polysaccharides (NSP) would include all the carbohydrate fractions and types of dietary fiber that are non-starch: soluble and insoluble celluloses, hemicelluloses, pectins, gums, mucilages, ß-glucans and noncellulosic polysaccharides (Bingham, 1987; Lanza and Butrum, 1986). Resistant starch (RS), by definition, is resistant to digestion by human digestive enzymes, with a

substantial amount reaching the colon undigested and is available for fermentation by the colonic microflora (Spiller, 1991).

Originally, a crude fiber analysis was adopted by the Association of Official Analytical Chemists (AOAC) in 1936 to determine the amount of lignin and cellulose in plant cell walls. This procedure involves extraction of the DF with strong acids and alkalies and severely underestimates the total fiber value by destroying part of the cellulose and lignin and completely destroying all the hemicellulose, gums and pectins present. It is generally agreed that the term, crude fiber, and the analyzation method should not be used when defining DF (Southgate et al., 1993). A gravimetric, neutral detergent fiber (NDF) method developed by Goering and Van Soest (1970) uses detergents for fractioning cell wall constituents and thus recovers all cellulose, some hemicelluloses and lignin and gives a relative measure of total plant fiber. This method was improved upon by the inclusion of an enzymatic steps to remove any starch or protein which may still be present in the sample (Schaller, 1977). The method has been criticized because it is gravimetric; however, detailed chemical analysis of the NDF residue have verified that the gravimetric yield is generally comparable to the yield

obtained by chemical analysis (Marlett and Chesters, 1985). A similar procedure to the NDF method is the acid detergent fiber (ADF) method developed by Goering and Van Soest (1970) which leaves cellulose and lignin and probably some other acid-insoluble substances, such as cutin and silica, as residuals for measurement. When NDF and ADF analyses are performed on the same residue, the difference between the procedures is often used to estimate water-insoluble hemicelluloses. The ADF fraction may contain some pectin and hemicelluloses. The currently approved American Association of Cereal Chemists (AACC) procedure for determining fiber is an enzymatic NDF method (AACC method 32-20). The crude fiber technique is still the accepted AOAC method used today (AOAC Method 978.10 or 962.09, 1990); although the AOAC has given final action status to a gravimetric NDF method that uses three sequential enzymes: a heat stable α-amylase for starch gelatinization; a protease for protein hydrolysis; and a fungal α-amylase for starch hydrolysis, with approval pending (AOAC, 1990).

The NDF procedure fails to include water-soluble fibers (the fiber material which dissolves in water at 100°C and precipitates in alcohol) which recently have been indicated as physiologically important (Mongeau and Brassard, 1986). Supplementing the enzyme-NDF procedure with a simple

procedure of soluble dietary fiber (SDF) has been reported to provide a practical method for rapid determination of total dietary fiber (TDF) in human foodstuffs (Mongeau and Brassard, 1990). Other analyzation techniques have been developed including fiber determination by near-infrared reflectance spectroscopy (Baker, 1983; McDonald, 1986) and a simple calculable method for estimating neutral detergent fiber content of typical daily menus (Johnson and Marlett, 1986).

TABLE 1

COMPONENTS OF DIETARY FIBER

Water Soluble Substances	-	Pectins
	-	Gums and Mucilages
	-	Some Hemicelluloses
Water Insoluble Substances	-	Cellulose
	-	Some Hemicelluloses
	-	Lignin
	-	Cutin, Silica and Ash
Other	-	Phytic Acid

Selvendran RR. The plant cell wall as a source of dietary fiber: chemistry and structure. Am J Clin Nutr 1984;39:320-337.

II. Components of Plant Cell Walls and Dietary Fiber

A. General

Plant cells are enclosed by a wall which is responsible for protecting cellular contents and are held together by the middle lamella composed of pectin (Selvendran, 1984; Campbell et al., 1979). The primary cell wall is a mix of cellulose, hemicellulose and some pectin. Depending on the maturity and species of the plant, a secondary cell wall may or may not be formed. Within this secondary wall cellulose, hemicelluloses and lignin may be found. Lignin acts to stiffen plant cell walls and render them less flexible and more wood like. Lignin is not often a concern of the human nutritionist because highly lignified vegetables are often not palatable and therefore not often consumed by humans (Campbell et al., 1979).

Dietary fibers can be classified into two categories: those components which are soluble in water and those components insoluble in water. The water soluble components include a portion of the hemicellulose, storage polysaccharides, mucilages, pectins and gums. Water insoluble components include cellulose, lignin and a portion of the hemicelluloses (Anderson and Chen, 1979) and is roughly equivalent to the term - nonstarch polysaccharide

(NSP) (Lanza and Butrum, 1986; Southgate, 1992).

B. Water Soluble Components of Dietary Fiber

1. Pectic Substances

This group of cell wall polysaccharides consists of pectic acid, pectinic acid, pectin and protopectin (Campbell et al., 1979). These substances are characterized by polyuronide molecules composed of galacturoinc acid units combined by alpha-1,4 glycosidic linkages. Pectin is highly water soluble and capable of salt formation by the presence of many carboxyl groups making it acidic and capable of reacting with metal ions to form salts. This property of being an ion exchange resin could be responsible for the binding of certain minerals (Kelsay, 1978). Commercially produced pectin forms gels in the presence of acids and sugar and is mainly in the form of pectinic acid. Being water soluble, pectin is often lost due to analysis procedures requiring boiling or extracting with acids or alkalis.

2. Gums and Mucilaginous Compounds

Gums are complex highly branched polysaccharides that swell in water to form gels or sticky solutions (Raven et al., 1976). Gums are produced by a process referred to as

gummosis (Lewis, 1978). Gummosis occurs in specially formed cell complexes or in ordinary tissue. Often caused by diseases, insect damage or physical injury, the process is the formation of cells and then the disintegration of the cell wall. It begins in the primary wall and proceeds inward to the innermost lamella. The cavity formed is then filled with gum.

Mucilages are gelatinous substances isolated from seaweed that is composed of very complex polyuronide proteinaceous matter and cellulose (Raven et al., 1976). Any plant substance that swells in water to form a slimy, colloid solution is considered a mucilageous compound (Southgate et al., 1993).

3. Hemicelluloses

This plant cell wall constituent is a very complex carbohydrate containing residues of various sugars in a variety of glycosidic linkages (Van Soest, 1978a). Sugars often identified with hemicellulose are arabinose, xylose, xyloglucose, galactose and mannose (Campbell et al., 1979). Not very similar chemically to cellulose, hemicellulose is somewhat chemically or physically similar to pectins, who together form the matrix of the plant cell wall in which are enmeshed cellulose fibers (Robertson, 1976; Southgate et

al., 1993). Hemicelluloses are insoluble in water alone but are soluble in dilute alkali after delignification (Selevendran, 1984; Campbell et al., 1979; Robertson, 1976). As with pectin, hemicellulose can demonstrate ion-exchange capabilities as a consequence of the presence of uronic acids; D-glucuronic, D-galacturonic and D-mannuronic acids (Robertson, 1976). Of importance, with respect to oat bran, are the beta-D-glucans; water soluble hemicelluloses containing both beta-(1-4)- and beta-(1-3)-linked D-glucosyl residues in a ratio of about 2.4:1 (Klopfenstein, 1988; Englyst et al., 1991). It is this soluble fiber which is thought to be a major factor responsible for oat fiber's ability to lower serum cholesterol levels (Vollendorf and Marlett, 1991). Studies by Wood et al. (1989) indicate that the soluble dietary fiber of oats is physiologically active and may be a beneficial component of oat products in the human diet. Beta-glucans have lower molecular weights than cellulose and tend to form sticky, viscous solutions when mixed with water (Wood, 1986; Klopfenstein, 1988; Wood et al., 1989). Glucans may impair nutritional status by interfering with the absorption of nutrients from the diet especially mineral bioavailability (Klopfenstein, 1988). The American Association of Cereal Chemists recently adopted a definition of oat bran which specifies a minimum ß-glucan

content of 5.5% and a total dietary fiber content of at least 16%, both on a dry weight basis, with one third of this dietary fiber being soluble fiber (Vollendorf and Marlett, 1991).

4. Phytate (Phytic acid)

Phytic acid (myo-inositol 1,2,3,4,5,6-hexakis inositol dihydrogen phosphate) is a phosphorus storage compound that is found in all cereals, many legumes and nuts and in a few tubers and roots (Davies, 1979). It is known to be digestible in some animals and proposed to be in man, although the extent of its digestibility in man remains to be determined (Van Soest, 1978b). Phytate is found in the soluble fraction of dietary fiber and is the principle soluble chelating agent (Rendleman, 1982; Frølich and Nyman, 1988). The ability of phytic acid to complex minerals has been known for some time. The proposed mechanism by which phytic acid impairs mineral utilization is due to its ability to bind minerals at the pH observed in the intestine to form an insoluble mineral-phytate complex which is unavailable for absorption (Davies, 1979). Research has mainly focused on phytic acids' effects on divalent ions of zinc, calcium, iron, magnesium and copper. A recent review of current literature by Munoz and Harland (1993) concludes

that in experimental human studies phytate was associated with decreased utilization of calcium, copper, iron, magnesium, phosphorus and zinc.

Researchers are divided on the topic of phytic acid vs. DF as the factor most limiting mineral absorption. Reinhold et al. (1973) concluded that deficiencies in zinc and calcium among Iranian villagers was due to high consumptions of phytic acid. But in 1976, Reinhold et al. concluded from studies involving two individuals that the fiber of wheat was largely responsible for calcium, magnesium and zinc binding. Other studies have been done supporting both conclusions leaving the question unresolved.

C. Water Insoluble Components of Dietary Fiber

1. Cellulose

Cellulose is a polysaccharide composed of glucose units combined with beta-1-4 glucosidic linkages and is the most abundant molecule in nature (Van Soest, 1978a; Robertson, 1976; Lanza and Butrum, 1986). Its main purpose is structural and it is the only true "fibrous" component in the plant cell wall. Cellulose molecules are combined in orderly linear crystalline arrangements which form microfibrils. It is highly insoluble and indigestible due to this molecular arrangement and beta linkages.

Cellulose content, as determined by the crude fiber method considerably underestimates the true amount of food or feed cellulose. Robertson (1976) reports that the crude fiber method destroys from 20 to 50% of the cellulose present. Goering and Van Soest (1970) developed a rapid procedure for ligno-cellulose determination using the acid detergent system. It recovers all the cellulose and lignin while removing more than 95% of the hemicellulose.

Studies involving cellulose as a source of DF often use refined or purified cellulose which has been chemically altered which drastically changes its biological properties. Slavin, Brauer and Marlett (1981) report that the digestibility of refined cellulose is minimal.

2. Lignin and Other Phenolic Compounds

The term lignin covers a group of related compounds which are noncarbohydrate constituents of plant cell walls (Campbell et al., 1979; Lanza and Butrum, 1986). True lignin is polyphenolic in structure, has a condensed non-hydrolyzable structure and is indigestible (Van Soest, 1978a). Lignin is laid down in plant cell walls to provide increased strength in the existing structural polysaccharides (Van Soest, 1978b). By this association, lignin acts to depress their digestibility. Lignin is

deposited in the plant cell wall as the plant matures, eventually providing a "woody" texture to the plant (Selvendran, 1976).

The heating of foodstuffs at temperatures above 50° to 55°C frequently results in the production of artifact lignin via the Maillard reaction (Goering and Van Soest, 1970; Van Soest, 1978b). The Maillard reaction or noneyzmatic browning occurs when carbohydrates and proteins condense to form an insoluble polymer.

3. Cutin, Silica and Ash

Cutin is a common waxy substance found in the outer cell walls of higher plants. It is a mix of complex macromolecules of long chain fatty acids and polyhydroly derivatives which are crosslinked through ester bonds to form a large interlinking matrix relatively impervious to water and gases (Raven et al., 1976; Bailey et al., 1978). The relation of cutin to the nutritive value of the other plant constituents is not well understood.

Silicon is deposited in plant cell walls as silica. Silica uptake depends on the species of plant and on soil concentrations. It appears that this element is an integral and essential constituent of polyuronides (pectins and micropolysaccharides) (Jones, 1978). Silica appears to

promote resistance to fungi and insect attack by contributing to the rigidity of the plant stem.

Ash consists of those metal cations that are intimately associated with the structural polysaccharides in the cell wall. In principle, the binding of a metal cation by the organic constituents of plant cell walls, and possibly to silica, could be expected to reduce availability of the cation for intestinal absorption (Jones, 1978). Sizeable amounts of some minerals are bound and not available for absorption. Reinhold et al. (1973) surveyed Iranian villagers and found several mineral deficiencies in spite of high intakes. They concluded that phytic acid and fiber were binding these minerals rendering them unavailable.

Ash may also express mineral-mineral interactions with respect to absorption. Interaction between zinc and copper or calcium and magnesium is mainly due to their very similar physical and chemical properties (Davies, 1979). Their similar properties enable them to interchange with one another and compete for intestinal binding sites.

D. Oat and Oat Bran Fiber

Oats belong to the genus Avena with over 500 different species identified. As a cereal grain, oat ranks 5th among the various cereal grains cultivated today (Table 2). The most common grown specie of oats is A. sativa.

Cultivated as early as 386 AD in China, oats appear to be a more recently developed grain as compared to rye or wheat. Oats are annual plants cultivated and harvested in much the same manner as wheat. Oats grow well in temperate climate regions around the world. They are less resistant to cold weather than wheat and are normally planted in the spring of the year. A. sativa or common oats is grown in cool, moist regions of northern United States and southern Canada (Baum, 1977; Murphy and Hoffman, 1992). A. sterilis or red oats is more adapted to drier, warmer regions of the southern U.S.. Leading oat producing countries are the former Soviet Union, USA, and Canada (Murphy and Hoffman, 1992; Schrickel, 1986).

The highest quality oats are used for human consumption as oatmeal or manufactured into rolled oats and various cereal products. However, the main use of oats is as an animal feed. Either way, when received for processing the oat grains are cleaned and heated slightly to preserve vitamins and minerals (Siebert, 1987). The hulls, which are

of scant nutrient quality, are separated from the groats during the milling process. Groats are used for the production of various oat products. Rolled or old-fashioned oats are simply whole groats that have been slightly steamed which are passed through rollers to flatten the groat and increase the surface area for faster cooking. Quick-cooking oats are made by cutting the groat into several pieces, then rolling the segments quite thinly. Instant oats (cook-in-the-bowl) are made from groats which have been further steamed and compressed into flakes (Burnette et al., 1992).

The nutrient content of oats depends on the soil concentrations of nutrients. Oats contain one-third more protein than wheat and nearly four times as much fat and less starch (Lockhart and Hurt, 1986; Peterson, 1992). Analyses of the dry matter of whole oats is presented in Table 3.

In contrast to other cereal grains, little information is available in the literature which pertains specifically to the sugars and NSP content of oat bran (MacArthur-Grant, 1986). DF content of oats was determined by Van Soest (1978b) to contain:

6.9%	cell wall
1.1%	lignin
1.0%	cellulose
4.8%	hemicellulose
2.0%	crude fiber

The dietary fiber of oats is a mixture of soluble and insoluble fractions with a high soluble fraction relative to other cereals (Peterson, 1993). The soluble fiber fraction appears to be mainly β-glucans, water soluble hemicelluloses containing both beta-(1-4)- and beta-(1-3)-linked D-glucosyl residues in a ratio of about 2.4:1 (Klopfenstein, 1988). The irregular configuration makes these polymers partially water soluble and functionally different from cellulose in the human digestive system (Peterson, 1993). It is this soluble fiber which is thought to be a major factor responsible for oat fiber's ability to lower serum cholesterol levels (Vollendorf and Marlett, 1991). Studies by Wood et al. (1989) indicate that the soluble dietary fiber of oats is physiologically active and may be a beneficial component of oat products in the human diet. Beta-glucans have lower molecular weights than cellulose and tend to form sticky, viscous solutions when mixed with water (Wood, 1986; Klopfenstein, 1988; Wood et al., 1989). Oat β-glucans are enriched in the bran fraction and therefore are not lost during the milling process since most oats for human consumption are essentially a flattened, whole grain.

Mineral content of oats is dependent on the concentration of the minerals in the soil, environmental conditions and agricultural practices (Lockhart and Hurt,

1986). Mahoney (1982) reported on the mineral content of over 200 commercially produced cereal products. McKechnie (1983) presented data on the ash and nutrient content of milled oats in his paper on oat products and their uses in foods (Table 4). Oats appear to contain ample amounts of calcium, magnesium, zinc, and copper but Peterson (1992) reports that variations in mineral content do exist. He concluded from reviewing several studies that the concentrations of phosphorus, potassium, and magnesium were relatively stable, whereas concentrations of the other minerals (Ca, Cu, Fe, Mn, Zn) were more variable. Exactly how much of these minerals are available for utilization by man is questionable. The DF content is relatively high possibly leading to increased binding of the available minerals rendering them unabsorbable. Frϕlich and Nyman (1988) determined the mineral content obtained from four milling fractions; course bran, fine bran, bran flour and white flour. The course bran fraction contained more of each mineral than any other fraction with a greater proportion of each mineral associated with the soluble fiber components of oat bran, but it could not be determined if the minerals were bound to β-glucan or to phytic acid which is also found in the soluble fiber fraction.

The oat products used in this study were an unprocessed

oat bran cooked cereal marketed as Mother's Oat Bran[1] and a processed ready-to-eat (RTE) cereal of experimental nature[1]. A low fiber cooked cereal, Cream of Wheat[2], was used as the control diet cereal.

[1] Quaker Oats Company, Barrington, IL.

[2]Nabisco Brands, Inc., East Hanover, NJ.

TABLE 2

CEREAL GRAIN RANKING ON HUMAN CONSUMPTION

1	Wheat
2	Corn
3	Rice
4	Barley
5	Oats
6	Millet
7	Rye

Baum, B.R.. Oats: Wild and Cultivated. Canada Department of Agriculture Research Branch. Ottawa, Ontario, canada. 1977, Monograph #14.

TABLE 3

ANALYSIS OF DRY MATTER IN WHOLE OATS*

Nitrogen Free Extract	67.0%
Protein	13.2%
Fat	5.6%
Fiber-Crude	10.8%
Ash	3.4%

* based on 100 gram sample

Thornton, H.J.. The History of the Quaker Oats Company. The University of Chicago Press. Chicago, IL. 1933.

TABLE 4

PROXIMATE ANALYSIS OF MILLED OAT PRODUCTS*

Carbohydrate	65.0%
Protein (Dry Basis)	17.0%
Fat	6.5%
Crude Fiber	1.3%
Ash	1.7%
Moisture	9.5%
Calories/100 g	380.0

* based on 100 gram sample

McKechnie, R.. Oat Products in Bakery Foods. Cereal Food World. 1983. 28:635-637.

III. Minerals

Minerals as presented here, refers to the elements in their simple inorganic form (Krause and Mahan, 1976). There are 21 known minerals that are essential to growth and life in man. The minerals present are mainly in an ionic form: metals forming positive ions and non-metals forming negative ions.

Minerals perform many functions in the body. They function in the skeletal system with the formation and maintenance of bones and teeth. The balance of mineral ions in body fluids regulates the metabolism of many enzymes, maintains acid-base balance and osmotic pressure, facilitates membrane transfer of essential compounds, maintains nerve and muscle irritability and in some cases mineral ions are building constituents of body tissue (Krause and Mahan, 1976).

Essential minerals are divided into macro-minerals and micro-minerals depending on the concentration of the mineral in the body. Macro-minerals are required in amounts of 100 mg/day or more. Calcium and magnesium are examples. Zinc and copper are two essential micro-minerals required in only a few milligrams per day.

This investigation looked at the minerals calcium, magnesium, zinc, and copper using human metabolic balance

methods to determine if unprocessed or processed oat bran had any effect on these minerals' balance or apparent retention or plasma levels in adult males.

A. Calcium

The adult human body contains an average of 1200 grams of calcium, making it the most abundant mineral present in the body (Krause and Mahan, 1976; NRC, 1989). Of the calcium present, 99% is found in the hard tissues (bone and teeth) with 1% located in the extracellular fluids and in cells (NRC, 1989; Pike and Brown, 1975). The U.S. Recommended Dietary Allowance (RDA) for calcium, based on balance studies, is currently set at 800 mg/day for adults aged 25 to 50 (NRC, 1989). Calcium intakes for normal healthy adult Americans have been studied by various investigators. Pennington et al. (1986) reported on the estimated daily mineral intakes of the US population using data from the Selected Minerals in Foods Survey, a portion of the Food and Drug Administration's (FDA) yearly Total Diet Study. They found that intake levels of calcium were lower than the 1980 RDA set levels for most age-sex groups. Further research by Pennington et al. (1989), also using the FDA's Selected Minerals in Foods Survey portion of the Total Diet Study for 1982 to 1986 found that intakes of calcium

were below the 1980 RDA set levels for three or more of the age-sex groups. Morgan et al. (1985) reviewed data from the USDA's 1977-1978 Nationwide Food Consumption Survey and found that a majority of the US population consumed less than the 1980 RDA levels. Block and Subar (1992) reported estimates of nutrient intakes from the 1987 National Health Interview Survey and found that for 5 age groups surveyed, men appeared to consume nearly the RDA set level (800 mg/day) for calcium while women in these same age groups consumed well below the RDA set level. Normal plasma calcium concentration ranges from 9 to 11 mg/dl (Martin et al., 1981). Plasma concentration and dietary intake appear not to be correlated (Kant et al., 1989).

Calcium has many functions in the body. It acts structurally in bones and teeth. Calcium present in the skeleton is held in the form of deposits of calcium phosphates, mainly hydroxyapatite, within a soft, fibrous, organic matrix (NRC, 1989). Calcium affects nerve and muscle function, hormonal actions, blood clotting and cellular motility (NRC, 1989; Martin et al., 1981). A tight control on plasma calcium levels and therefore on the metabolism of bone depends on the ability of parathyroid hormone to mobilize calcium from the bone when plasma calcium levels fall, and on the inhibition of bone

mobilization by thyrocalcitonin when plasma calcium levels rise (Martin et al., 1981).

Calcium absorption takes place by active transport involving a calcium binding protein and by passive diffusion in the duodenum (Krause and Mahan, 1979; Martin et al., 1981). Of the calcium ingested only about 15 to 20% is absorbed by the adult. The efficiency of absorption is increased during periods of high physiological need. Absorption is affected by gastric pH, the presence of adequate vitamin D, the presence of oxalates, phosphates and phytates which form insoluble calcium salts and certain dietary fiber fractions (Krause and Mahan, 1979; Martin et al., 1981; NRC, 1989). Other dietary minerals may compete with calcium for active transport. Smith (1988) reviewed the effects of calcium and other trace mineral interactions, mainly zinc and iron. It is apparent that some mineral interactions do exist and must be taken into account when considering fortification programs. An in-vivo study done by Roth-Bassell and Clydesdale (1991) using rat brush border membrane vesicles showed that at ratios of 1:1, based on the RDA, none of the studied minerals (Zn, Mg, or Fe) were found to significantly decrease calcium uptake, but at higher RDA ratios magnesium and zinc both inhibited calcium uptake (ratio of 3:1 Mg:Ca and 10:1 Zn:Ca).

Excretion of calcium is through urine, feces, dermal losses and sweat. While an individuals' urinary calcium excretion is relatively constant over a wide range of intakes, fecal losses correlate with daily calcium intakes (Krause and Mahan, 1976). A person is said to be in positive calcium balance when intake exceeds excretion and calcium is retained by the body. Good food sources of calcium include dairy products, dark green leafy vegetables, sardines and salmon, lime-processed tortillas and calcium fortified foods (NRC, 1989).

B. Magnesium

The adult human body contains approximately 20 to 30 grams of magnesium of which ~59% is found complexed in hard tissue, 40% in muscles with the remainder in soft tissues and body fluids (NRC, 1989). The function of magnesium in hard tissues is not fully understood. Within the cells of soft tissues the concentration of magnesium is greater than any other mineral except potassium. Magnesium is required for cellular respiration, especially in oxidative phosphorylation leading to adenosine triphosphate (ATP) formation (Martin et al., 1981). Magnesium is essential for the production and transfer of energy for protein and nucleic acid synthesis, for contractility of muscle and

excitability in nerves. It is known that magnesium serves as a cofactor in numerous enzyme systems, particularly those requiring thiamin pyrophosphate (TPP). Overall, magnesium is important for > 300 different enzyme systems (Wester, 1987).

Absorption of magnesium is through active transport in the small intestine, both in the upper and lower parts (Wester, 1987). Magnesium competes with calcium for carrier sites during active transport. Approximately 50% of the ingested magnesium is absorbed (NRC, 1989). Absorption of magnesium varies inversely with intake: when intake is low the absorption rate is high and vice versa. The kidney is the major excretory pathway for absorbed magnesium. Urinary magnesium remains relatively constant due to efficient renal reabsorption (Wester, 1987). Normal plasma magnesium concentration ranges from 2 to 3 mg/dl (Martin et al., 1981) or 0.65 to 1.0 mM (NRC, 1989) and is believed to be regulated primarily by the kidney although there is no known homeostatic system regulating serum magnesium (Wester, 1987). Plasma concentration and dietary intake appear not to be correlated (Kant et al., 1989). Estimates of daily intakes of magnesium indicate adult Americans consume less than the 1980 RDA level of 300 mg/day for females and 350 mg/day for males (Pennington et al., 1986; Pennington et

al., 1989; Morgan et al., 1985). Magnesium absorption appears to be possibly altered by mineral competition for intestinal binding sites and by the presence of dietary fiber and/or phytates.

Good food sources of magnesium include meats and seafoods, whole seeds such as nuts, legumes and unmilled grains (NRC, 1989). The chlorophyll molecule has one atom of magnesium incorporated into it making dark green leafy vegetables another good food source. Among beverages, coffee, tea and cocoa are rich sources of magnesium (Wester, 1987). In hard-water regions, waterborne magnesium may contribute 9-27% of daily intake (Wester, 1987). The current RDA for magnesium is 350 mg/day for adult males and 280 mg/day for adult females (NRC, 1989). These requirements are based on magnesiums' function in ATP formation with males requiring more due to increased energy needs.

C. Zinc

Zinc is a trace mineral essential for life and growth in man. The adult human contains approximately 2 to 3 grams of zinc with the highest concentration found in the choroid of the eye, the male reproductive organs, liver, and voluntary muscle (Pike and Brown, 1975). Bone contains much

of the body's zinc but these stores are not readily available for equilibrium of body fluids. Normal plasma zinc concentrations range from 60 to 100 μg/dl (Martin et al., 1981). Plasma concentration and dietary intake of zinc appear not to be correlated (Kant et al., 1989). Plasma zinc concentrations are affected by a number of factors other than dietary intake such as infection, acute stress, pregnancy and oral contraceptive use. In addition, plasma zinc levels are subject to diurnal variation and influenced by the fasting status of the subject (Pilch and Senti, 1985).

Zinc functions essentially in at least 20 different enzyme systems. It acts as a cofactor in the synthesis of DNA and RNA. This function is especially important in systems with a high rate of cellular turnover as in the gastrointestinal tract and taste buds (Martin et al., 1981). Zinc aids in the mobilization of vitamin A from the liver to maintain normal blood vitamin A levels. It is also thought that zinc enhances the action of certain hormones but no exact mechanism is known.

Absorption of zinc is in the upper small intestine and is apparently promoted by a zinc-binding protein secreted by the pancreas (Martin et al., 1981). After absorption, zinc is transferred to albumin for transport in the blood. Zinc

is excreted through many routes, principally through feces but also through urine, sweat and the sloughing of mucosal cells.

A true zinc deficiency in humans was first described by Prasad et al. (1963) in areas of Iran and the Middle East where the diet consists mainly of unleavened wholemeal bread. This bread contains considerable amounts of phytate and dietary fiber which are known to bind zinc rendering it unabsorbable. Symptoms of zinc deficiency include poor growth, loss of appetite, hypogonadism in males and a loss of sense of smell and taste. The teratogenic effect of zinc deficiency in humans has not been demonstrated; however, there is evidence of this effect in animals (Krause and Mahan, 1976).

The current RDA for zinc is set at 15 mg/day for adult males and 12 mg/day for adult females (NRC, 1989). There has been some concern about this RDA level since most North Americans consume on the average of 12 mg zinc per day in a mixed U.S. diet (NRC, 1989). A study by Gibson and Scythes (1982) found that women consuming self-selected diets had a mean zinc intake of 10.1 mg/day. Pennington et al. (1986) reported on the estimated daily mineral intakes of the U.S. population using data from the Selected Minerals in Foods Survey a portion of the Food and Drug Administration's (FDA)

yearly Total Diet Study. They found that intake levels of zinc were lower than the 1980 RDA levels for most age-sex groups. Reported zinc intakes as a percentage of the 1980 RDA were higher for males compared to females in the same age-sex groups. But further research by Pennington et al. (1989), also using the FDA's Selected Minerals in Foods Survey portion of the Total Diet Study for 1982 to 1986, found that intakes of zinc exceeded the 1980 RDA levels for infants, teenage boys and adult men while intakes were below the RDA levels for teenage, adult and older females. Estimates of zinc intakes of young women using dietary data from the NHANES II study were calculated to be 8.11 ± 0.14 mg/day (Murphy and Calloway, 1986). Retzlaff et al. (1991) investigated the nutritional adequacy of diets based on guidelines recommended by the 1987 National Cholesterol Education Program (NCEP) to promote dietary modification as the first line of treatment for hypercholesterolemia. They reported a mean zinc intake of 80% of the RDA at baseline but observed a decrease to 75% of RDA when intakes of meat, fish and poultry were limited to 85g/day. Meats and other animal products appear to be good sources of zinc. Most of the zinc consumed in plant products comes from cereals but bioavailability varies with a number of factors such as phytate, dietary fiber and protein content.

D. Copper

An adult human body contains on the average of 80 to 120 mg of elemental copper. The highest concentrations of copper are found in the liver, brain, heart, and kidney (Krause and Mahan, 1976; Martin et al., 1981). The absolute body concentration of copper is seen to remain remarkably steady throughout adult life (NRC, 1989). In blood, copper appears equally distributed between plasma and erythrocytes (RBC's). Normal plasma levels range from 90 to 140 μg/dl (Krause and Mahan, 1976). Approximately 90% of the copper in plasma is bound to ceruloplasmin, a glycoprotein. Ceruloplasmin is not a copper transport protein but is a copper-dependent ferroxidase that contains 6 to 8 atoms of copper (Pike and Brown, 1975; Martin et al., 1981). Ceruloplasmin is strongly influenced by hormonal changes or inflammation thus limiting its usefulness as an indicator (NRC, 1989).

Copper is important in various enzymatic processes throughout the body, especially oxidative reactions. A number of important copper containing proteins and enzymes have been identified; some of which are essential for proper iron utilization. Ceruloplasmin, for example, is necessary for oxidation of ferrus iron (Fe^{++}) to ferric iron (Fe^{+++}).

Dietary copper deficiency is not known to occur in adults under normal circumstances but has been diagnosed in children (Pike and Brown, 1975). The current estimated safe and adequate daily dietary intake (ESADDI) recommends 1.5 to 3 mg/day for adults (NRC, 1989). As with other minerals, there has been concern that the average North American diet does not furnish this amount. Surveys of various diets have indicated that actual intakes may be as low as 1 mg/day (Sandstead, 1982). Copper consumption in females on self-selected diets gave a mean intake of 1.9 mg/day which is within the current estimated safe daily dietary intake proposed by the National Research Council (Gibson and Scythes, 1982). Pennington et al. (1986) reported that for all age-sex groups studied, subjects consumed below the 1980 estimated safe daily dietary intake levels of copper. Further study by Pennington et al. (1989) indicated copper intakes for adult males and females averaged about 1.2 and 0.9 mg/day respectively, based on data from the Total Diet Study from 1982 to 1986. Estimates of copper intakes of young women using dietary data from the NHANES II study were calculated to be 1.16 ± 0.02 mg/day (Murphy and Calloway, 1986). Plasma concentration and dietary intake of copper appears not to be correlated (Kant et al., 1989).

Absorption of copper in the gastrointestinal tract

requires a specific mechanism because of the highly insoluble nature of cupric ions (Cu^{++}) (Gibson and Scythes, 1982). In the intestinal mucosal cell, copper is associated with a low-molecular-weight, metal-binding protein, metallothionein. From here, copper is bound over to amino acids and serum albumin which travels through the bloodstream to the liver. At the liver, copper is processed through two routes: biliary excretion into the gastrointestinal tract and through incorporation into ceruloplasmin (Martin et al., 1981). Copper homeostasis is maintained almost exclusively by biliary excretion. Trace amounts of copper are found in urine. This laboratory has previously determined urinary copper to be negligible when considering excretion (Taper et al., 1980).

The richest sources of copper are legumes, wheat bran cereals, organ meat especially liver, shellfish, and grains with a variable amount found in drinking water (Krause and Mahan, 1976; Lurie et al., 1989). When considering copper bioavailability, Sandstead (1982) reported that the ratio of zinc to copper affected the absorption of copper. It is thought that since zinc and copper are similar metals the chemical characteristics of zinc impairs copper absorption and metabolism. Copper balance in human subjects has been

reported to be improved or lowered with fiber sources added to the diet (Turnlund, 1982) but further research is needed to more accurately describe any effects dietary fiber has on copper availability.

IV. Factors Affecting Mineral Absorption

A. pH

A proper pH is necessary for absorption of divalent minerals such as calcium, magnesium, zinc, and copper. Gastric juices are highly acidic with a pH of about 1.0 (Martin et al., 1981). This acidity is due to parietal cell activation and secretion of hydrochloric acid into the stomach. As the stomachs' contents become acidic, food begins to digest. Minerals change from insoluble, complexed forms into separate cations with positive charges when exposed to this acidic environment. To be absorbed, minerals must be released from complexes in food and made soluble (Champagne, 1988). No mineral absorption, as far as we know, occurs in the stomach. As chyme leaves the stomach and enters the duodenum it is neutralized and buffered to a pH of 5.5 to 7.5 by pancreatic and biliary secretion (Martin et al., 1981). Most mineral absorption occurs in the duodenum where the pH of chyme is changing from highly

acidic to almost neutral. It appears that an acidic pH is necessary for normal calcium absorption (Krause and Mahan, 1976). Since magnesium and calcium compete for carrier proteins for absorption it can be postulated that an acidic environment is possibly necessary for normal magnesium absorption as well.

Dietary fiber is also affected by the pH of the gastrointestinal juices. Dietary fiber is a rich source of most minerals but these minerals only become soluble and free for absorption under certain pH conditions. Champagne (1988) states that at pH values encountered in the gastrointestinal tract, phytic acid would be strongly negatively charged and have immense potential for binding positively charged cations. At low pH values the minerals are generally only weakly bound by food matrixes. Rendleman (1982) showed through in-vitro studies that calcium binding by white bread and components of bran increased with a decrease in pH from 8.3 to 5.5. This would indicate that as digesta becomes neutralized, the binding capacity of dietary fiber lessens, releasing calcium for intestinal absorption. Similar results to these were found by Camire and Clydesdale (1981) and by Schweizer et al. (1984) through in-vitro experimentation.

In-vitro studies on zinc and copper indicate that for

these two minerals an increase in pH, as occurs in the duodenum, decreases the minerals' solubility from wheat bran and wholemeal bread (Reilly, 1977). It was found that at a pH of 4.3 zinc is much more soluble than copper with approximately 80% dissolved into the media. Reilly concluded that although wheat bran and wholemeal bread are rich sources of zinc and copper, at the pH in the duodenum only small quantities of these minerals are released for absorption. Similar results have been found through in-vitro experimentation by Ismail-Beigi, Faraji and Reinhold (1977), Camire and Clydesdale (1981), and Rendleman and Grobe (1982). A more recent study by Lyon (1984) using in-vitro methods on various cereal products of varying phytate content produced evidence that at an increase in pH to 7.0 zinc binding was at an optimum while calcium and magnesium binding were very low. Mod et al., (1982) investigated the in-vitro interaction of rice hemicellulose with trace minerals and their release by digestive enzymes (hemicellulase, pepsin and trypsin) and pH. Both enzymes and changes in pH released considerable amounts of bound minerals to differing degrees, suggesting that released minerals should be available for resorption in-vivo. An acid pH seemed to release more bound minerals.

Care must be taken when interpreting these in-vitro

results to in-vivo systems. In-vitro studies use purified environments and fibers which may or may not function the same in in-vivo systems. Other factors may be present in in-vivo systems which limit mineral bioavailability.

It appears that pH plays an important role in divalent mineral absorption. Acidic pH affects minerals by putting them in an ionic state while in solution. Cations are apparently recognized easily by carriers thus increasing mineral absorption. pH is also important in that it can bind or release minerals from dietary fiber and/or components of dietary fiber which may be present in the intestine with maximum binding just below neutral pH and minimal binding at acid pHs (Southgate, 1987).

B. Mineral-Mineral Interactions

Minerals themselves can affect other minerals' bioavailability. One type of interaction commonly observed between minerals arises because elements, through chemical nature or physical structure, act antagonistically to each other in biological systems (Davies, 1979). The interactions of zinc and copper can illustrate this point. The Zn^{++} and Cu^{++} ions have the same electronic structure and have similar tendencies to form stable complexes. In this

way the minerals can act as one another in functional sites due to this similar charge and physical properties (Davies, 1979). This may also be true of calcium and magnesium since competition between them for intestinal binding sites is known to occur (Roth-Bassell and Clydesdale, 1991; Wester, 1987). In a study by Spencer et al. (1987) pharmacological doses of zinc (140 mg/day as zinc sulfate) were added to a constant control diet which contained an average of 14.6 mg of zinc and containing a low calcium intake (230 mg/day) and to control diets containing a normal calcium intake (800 mg/day). It was found that during zinc supplementation, the intestinal absorption of calcium was significantly lower during a low calcium intake than in the control group. However, during the normal calcium intake (800 mg/day) the high zinc intake had no significant effect on the intestinal absorption of calcium. These studies indicate that a high zinc intake decreased the intestinal absorption of calcium during a low calcium intake but not during a normal calcium intake. In further studies of the effect of zinc on intestinal absorption of calcium in humans; Spencer, Norris and Osis (1992) looked to see if the same effect of zinc supplementation, but at a lower level than previously investigated, would also inhibit calcium absorption at a dietary level of 230 mg calcium/day. To study this, 100

mg/day of zinc sulfate was given daily during a low calcium intake of 230 mg/day. Results showed that the dose of zinc did not inhibit calcium absorption.

When considering mineral interactions the ratios of one mineral to another need also to be reviewed. Work done by Roth-Bassell and Clydesdale (1991) looked at the effects of adding increasing concentrations of zinc, magnesium, and iron on calcium uptake in rat, brush border membrane vesicles. They found that at ratios of 1:1, based on the RDA, none of these minerals were found to significantly decrease calcium uptake. Magnesium, however, inhibited calcium uptake at an RDA ratio of 3:1 (Mg:Ca) and at higher RDA ratios (Zn:Ca 10:1) zinc also decreased calcium uptake. Iron did not decrease calcium uptake but demonstrated an enhancing effect at high concentrations. Roth-Bassell and Clydesdale (1991) explained these effects by the actions of the ions individually. In other investigations, Smith (1988) evaluated four different sources of calcium (calcium carbonate, calcium phosphate, bone meal and synthetic calcium hydroxyapatite) with respect to their ability to interfere with iron absorption in rats. The different calcium sources were tested at ratios of 60:1 and 120:1 (Ca:Fe). The calcium and iron were dosed simultaneously or separated by intervals of approximately twenty minutes up to

a maximum of two hours. Although each calcium source inhibited iron absorption to some degree, the 120:1 ratios were all more inhibitory than the 60:1 ratios tested. Also, when dosed simultaneously, the inhibition was significantly evident. In addition, Smith also evaluated calcium-zinc interactions using the same calcium sources mentioned above. He found that calcium carbonate demonstrated a 25% inhibition of the zinc absorption as compared to zinc absorption when no calcium was present. All calcium sources were found to interfere significantly with zinc absorption at the 120:1 ratio level.

Dietary zinc is thought to affect copper absorption. Klevay has reported epidemiological and metabolic data that suggests an imbalance in zinc and copper intake is a major causative factor in cardiovascular disease (Klevay, 1975; Klevay et al., 1981). Work reported by Ritchey (1981) concludes that several dietary components are thought to affect zinc and copper nutriture.

Using an in-vitro method, Fischer, Giroux and L'Abbe' (1981) investigated the theory that zinc may have its antagonistic effect by competing with copper for common binding sites for absorption within the intestinal mucosa. Their conclusions suggest that in animal tissue, zinc exerts its antagonistic effect by inducing the synthesis of a

copper binding ligand which sequesters copper from the medium making it unavailable for absorption.

It is apparent that some divalent minerals act antagonistically toward each other with respect to absorption. Similar electronic charges and structures may signal intestinal binding.

C. Oxalates, Phosphates and Phytates

Oxalates, phosphates and phytates all complex divalent minerals into insoluble salts which are unabsorbable (Davies, 1979; Krause and Mahan, 1976). Rendleman (1982) reports from in-vitro studies using bran that water-soluble components are responsible for more than half the binding ability of bran, and the principle soluble chelating agent is phytate. Because of phytate's association with protein in cereal grains it is soluble and extractable at pHs above or below the isoelectric point of the major proteins in those grains (Oberleas, 1983).

Kelsay et al. (1988) investigated mineral balances of men fed a diet containing fiber from fruits and vegetables and oxalic acid from spinach. The investigators used a crossover design with subjects consuming diets for 6 weeks before switching dietary treatments. One diet contained about 25 g of neutral detergent fiber (NDF) in fruits and

vegetables and included 100 g of spinach as the oxalic acid source every other day, while the other diet was a low fiber diet that contained about 5 g NDF and the same amount of spinach as the first diet. It was found that mean mineral balances for calcium, magnesium, zinc, iron, and manganese were not significantly different due to diet. Copper balance was found to be significantly lower when the low fiber diet was consumed than when the diet containing fiber in fruits and vegetables was consumed. Differences were attributed to the fiber diet containing a naturally occurring higher level of copper.

Of the three (oxalates, phosphates and phytates), phytate has been well studied with respect to mineral binding. As reported earlier, research is divided on the topic of phytate vs. dietary fiber as the factor most limiting mineral absorption. In 1973, Reinhold et al. reported findings based on studies involving Iranian villagers. These people are known to exist mainly on tanok or shepards bread, an unleavened, wholemeal wheat bread. This bread is rich in phytate and dietary fiber and is thought to impair nutrient absorption. When subjects were placed on a metabolic diet of high quality, all mineral balances under study (Ca, Zn, P) went from negative to positive. Their results supported the theory that severe mineral depletion

exists in Iranian villagers and that the depletion in due to an over consumption of phytate in the form of tanok bread.

Turnlund et al. (1984) showed that the addition of phytate to a human diet, resulting in a phytate:zinc molar ratio of 15, greatly reduced zinc absorption and resulted in negative zinc balance in young men. While, Morris and Ellis (1983) found no difference in zinc balance when the phytate:zinc molar ratio of the diet was 1 or 10. Because of the possible deleterious effects of phytate:mineral molar ratios, it has been suggested that calculating the phytate:mineral molar ratios of diets may be helpful in identifying situations in which mineral malabsorption may occur (Munoz and Harland, 1993). Ellis et al. (1987) calculated the phytate:zinc and phytate x calcium:zinc millimolar ratios in self-selected diets of Americans, Asian Indians and Nepalese. With the phytate:zinc molar ratio used as an index (suggested critical value of 10 from animal studies), the results in their study suggest that phytate has little effect on the bioavailability of dietary zinc in most American omnivorous diets. A similar conclusion was found by Forbes et al. (1983) and Forbes et al. (1984) that phytate is not likely to exert a significant effect on zinc bioavailability to humans consuming usual and adequate diets. A similar study by Mason et al. (1990) looked at the

effect of moderately increased intakes of complex carbohydrates (cereals, vegetables and fruit) on iron and zinc metabolism in adult women. Fifteen subjects participated in a 14 week metabolic study where the intake of complex carbohydrates was increased from 20 g on the normal fiber diet to 30 g on the high fiber diet. Phytate:zinc ratios were 2.5 on the normal fiber diet and 3.5 on the high fiber diet. Although effects on bowel function were observed, the changed diet had no influence on iron or zinc retentions or on plasma mineral concentrations. Likewise, in a study on the effect of phytate and α-cellulose on copper absorption in young men, Turnlund et al. (1985) found that copper absorption was not significantly affected by high levels of either α-cellulose (0.5 g/kg body weight) or phytate (2.34 g sodium phytate) added to a basal diet.

A review of literature regarding the effects of phytate on mineral absorption indicates that phytate does exert some effect on mineral bioavailability with the most common outcome reflecting a decrease in calcium, magnesium, iron, zinc, copper, and phosphorus utilization (Munoz and Harland, 1993).

D. Dietary Fibers

1. Wheat

Wheat and wheat bran have been extensively studied with respect to mineral binding. As early as 1974, Heaton and Pomare (1974) revealed a significant decrease in plasma calcium levels in fourteen human subjects fed a diet supplemented with unprocessed wheat bran for 4 to 9 weeks. This hypocalcemic action of wheat bran was thought to be due to interference with intestinal calcium absorption. The concentration of calcium in blood was closely controlled and the decrease observed would indicate a negative balance. No fecal data on either dietary fiber or calcium was presented so actual balance was unknown. It must be noted that in contrast to normal bran, wheat bran for human nutrition must be free of starch and, to lower microbial counts, bran for human nutrition is subjected to high-temperature treatment (Becker et al., 1986). The effect of heat treatment may change the composition of the dietary fiber and thus complicate interpretation of results from studies using wheat bran. (See section entitled - Processing of Dietary Fiber.)

Reinhold et al. (1976) reported that a wholemeal wheat bread metabolic diet was responsible for negative balances

of calcium, magnesium, and zinc in two male subjects. The subjects experienced a significant increase in all fecal mineral excretions following the change from a white bread to a wheat bread diet. Negative balances for all minerals studied were seen only during the wheat bread feeding period. Fecal losses of calcium, magnesium, and zinc were correlated with fecal dry matter which in turn was directly proportional to fecal fiber excretion. It was apparent that fecal mineral excretion and ultimately, mineral balance was dependent on the wheat fiber consumption. To further this research, Ismail-Beigi, Faraji, and Reinhold (1977) performed several in-vitro experiments on wheat bread, wheat bran and components of dietary fiber with respect to zinc binding. Zinc binding was found to be highly pH dependent with lignin and hemicellulose fractions binding a greater percentage of zinc than cellulose or phytate.

Heaton, Manning, and Hartog (1976) found no effect on plasma calcium concentrations in young men upon changing from white to wholemeal bread over an extended time. Nineteen college students were recruited to consume along with their usual diets, 5 or more slices of a prepared wholemeal wheat bread for a length of 20 weeks. No attempt was made to determine calcium balance in the subjects but plasma calcium levels were monitored periodically throughout

the study. No significant changes in plasma calcium levels were seen, a finding contradictory to the results of Heaton and Pomare (1974).

An increase in dietary fiber consumption by adding about 26 g of a standard soft wheat bran was found to decrease zinc retention while improving copper retention in 5 male subjects under strict metabolic conditions in a study by Sandstead et al. (1978). Losses of zinc were mainly through increased fecal excretion. Urinary zinc remained relatively constant and unaffected by dietary fiber intake. Copper balance was improved, possibly because of an increased intake when fed the soft wheat bran. Copper balances were negative in 4 out of 5 subjects. Similar results were found by Sandberg et al. (1982) who added 16 g of wheat bran per day to the hospital diets of 8 ileostomy patients. When the wheat bran was fed, a significant decrease in zinc absorption was seen with no change in calcium or magnesium absorptions.

Increasing the amount of bran in bread from 9 to 22 g of NDF/day did not appear to affect the balances of calcium, copper, magnesium, zinc, or iron in young adult subjects (Van Dokkum et al., 1982). The increase in dietary fiber consumption resulted in a significant increased mineral intake, but also fecal excretion increased significantly.

Urinary excretion of calcium, zinc, iron, and copper remained unchanged, although a slight increase (non-significant) in urinary magnesium was seen. Similar results were also found by Andersson et al. (1983) when 6 subjects were fed metabolic diets containing wholemeal bread. The increase in dietary fiber consumption caused increased stool output but had no affect on calcium, zinc, and iron balances. They concluded that wheat bran is unlikely to exert a significant effect on mineral absorption in man in amounts customarily eaten as bread. But, in a study of the effect of wheat bran on bowel function and fecal calcium excretion in older adults, Balasubramanian et al. (1987) found that wheat bran's ability to regulate bowel function in apparently healthy older adults may be accompanied with increased fecal calcium losses. Seven healthy older adults participated in the study which was composed of a 10-day control period and two 10-day experimental periods, where, in addition to self-selected diets the subjects consumed a daily wheat bran supplement of 30 g. Apparent calcium absorption decreased significantly from the control period to the second bran period. The authors did not indicate if a time dependency was evident over the course of the study since the calcium absorption for period one was not significantly lower than the control period.

In a recent review of literature concerning wheat bran's possible effects on mineral utilization, Munoz and Harland (1993) found that the majority of animal studies involving wheat bran lowered mineral utilization for calcium, magnesium, iron, zinc, or copper. In a long term rat study by Shah et al. (1990) in which the effect of cereal brans on mineral metabolism was studied in a 7 month long experiment, overall conclusions indicated that absorption was affected by the level of that total mineral in the diet and not by the kind of fiber source. Rats were fed one of six fiber containing diets (cellulose, oat bran, hard red spring wheat, wheat bran, soft white wheat bran, corn bran or rat chow) at a fiber level of 4% or 14% of total dietary fiber for the course of the study. Two mineral balance periods were conducted during weeks 7 and 24, respectively. Results showed that the fractional absorption of calcium, phosphorus, and magnesium by the male or female rats was not significantly different. In most cases there was a decrease in absorption from week 7 to week 24 but this decrease was not significant.

It appears that wheat bran or a component of wheat fiber may negatively affect divalent mineral absorption. Fecal excretions of calcium, magnesium, zinc, and copper seem to be increased and this inturn negatively affects

retention. Urinary calcium, zinc and copper excretion is apparently not affected but several researchers reported increases in urinary magnesium excretion (Reinhold et al., 1976; Rendleman et al., 1982; Van Dokkum et al., 1982).

2. Cellulose

Cellulose is another component of dietary fiber that also has been extensively studied with respect to mineral binding. Cellulose is easily obtainable in purified form and is often used as a standard fiber in studies. In the purified form, cellulose is actually quite different chemically and structurally than naturally occurring cellulose making comparisons of the two difficult.

Platt and Clydesdale (1987) investigated the mineral binding characteristics of lignin, guar gum, cellulose, pectin, and neutral detergent fiber under simulated duodenal pH conditions in-vitro. To simulate the gastrointestinal digestive conditions they began studies at a pH of 2.0 (simulating gastric pH conditions) and increased the pH to 5.0 (simulating intestinal pH conditions). They found that lignin, but not cellulose or low methoxy pectin, was a potent binder of ferrous iron under pH conditions approximating those in the proximal intestine. Lignin had two binding sites for iron, zinc, and copper with the high

affinity sites binding these transition metals in this order: Fe > Cu > Zn, but with twice as much copper bound as either iron or zinc.

Added cellulose to the diets of laboratory animals has shown no adverse effects on mineral bioavailability (Tsai and Lei, 1979; van der Aar et al., 1983). Tsai and Lei (1979) added graded levels of cellulose to the diets of weanling rats along with two levels of zinc and copper. After 9 weeks on the diets the rats were sacrificed and several tissues removed for mineral analysis. Additions of cellulose appeared to have no effect on the distribution of zinc, copper or iron in tissues sensitive to dietary deficiencies. A slight decrease in serum zinc levels was seen as cellulose consumption increased but this reduction was not significant. In the study by van der Aar et al. (1983) different dietary fibers, among them cellulose, were studied for their effect on divalent cation absorption and utilization by female chicks. After 18 days on the diet the chicks were sacrificed and mineral determinations made. Cellulose had no effect on calcium, magnesium, zinc, or copper levels in serum or in mineral sensitive organs.

Studies involving humans have reached contradictory results as compared to those found by animal studies. Ismail-Beigi et al. (1977) added 10 g of cellulose to the

diets of 3 male subjects under metabolic conditions for 20 days. Fecal excretions of zinc and calcium were seen to increase significantly from baseline levels. Balances of both minerals became negative and plasma concentrations of both minerals decreased. Magnesium balance in 2 subjects became negative due to increased magnesium excretion via feces, although for all 3 subjects plasma concentrations of magnesium remained normal. Kelsay, Jacob and Prather (1979) studied the effects of a high fiber diet made up of fruits and vegetables on mineral balances in 12 male subjects. The fiber supplied by the fruits and vegetables contained more cellulose than would an equivalent amount of fiber from bran. The mean zinc and copper balances were found to be negative while on this diet. Mean fecal excretions of both minerals were increased on the high fiber diet. Further study by Kelsay et al. (1988) on the impact of variation of carbohydrate intake on mineral utilization by two groups of vegetarians (of Asian Indian and of American origin) compared with intakes of a group of nonvegetarians indicated that on self-selected diets the vegetarians had a significantly higher intake of crude fiber and neutral detergent fiber than the nonvegetarians. The American vegetarians had significantly higher intakes of iron, copper, and magnesium, than the other groups. Magnesium

intakes were higher for both groups of vegetarians over the nonvegetarians. For all the minerals studied (calcium, magnesium, iron, zinc, copper, and manganese) balances were negative for all three experimental groups. But calcium, iron, zinc, and copper balances were not significantly different among the groups. Magnesium and manganese balances were significantly more negative for the American vegetarians than for the two other groups. It was concluded by the authors that, in general, the higher percentage of carbohydrate intake or the higher level of fiber intake did not appear to affect mineral utilization by vegetarians. Intakes of minerals do vary when people are consuming self-selected diets and it is expected that balances in adults would fluctuate between positive and negative. Similar results were found by Behall et al. (1987) in adult men consuming diets with added refined fibers. A basal diet alone and with refined fiber added in one of four forms (cellulose, Na-carboxymethylcellulose, locust bean gum, or karaya gum) at 7.5 g fiber/1000 kcal was fed to eleven men for 4 weeks each. The adding of refined fibers to the basal diet did not significantly affect apparent mineral balance of calcium, magnesium, manganese, iron, copper, or zinc with the exception of a negative balance for manganese with carboxymethylcellulose.

In a recent review of literature concerning cellulose's possible effects on mineral utilization, Munoz and Harland (1993) found that the majority of human studies involving cellulose indicated lowered mineral utilization for calcium, magnesium, iron, zinc, copper, and phosphorous, while a few studies showed no effect of cellulose on mineral utilization by humans.

3. Hemicellulose

Hemicellulose is known to have ion exchange capabilities which may affect mineral binding principally due to the physical properties of cell wall materials to act as weak cation exchangers with the capacity to bind divalent ions (Southgate, 1987). A study by Kies, Fox, and Beshgetoor (1979) looked at the effects of feeding various levels of hemicellulose on zinc nutritional status of men. Twelve men were fed metabolic diets containing 4.2, 14.2, and 24.2 g of hemicellulose for periods of 14 days each. They found that as the amount of hemicellulose in the diets increased the amount of zinc excreted via the feces increased and resulted in a significantly negative zinc balance for the 24.2 g supplemented group. Urinary and blood serum zinc levels were unaffected by the increases in dietary hemicellulose. Similar results were also found by

Drews, Kies, and Fox (1979) when dietary fiber was fed to adolescent boys. Eight male subjects were fed a basal diet alone and with an additional 14.2 g of added cellulose, hemicellulose or pectin for 21 days. Zinc, magnesium, and copper balances were significantly lowered when the subjects consumed the hemicellulose. Small, non-significant changes in urinary excretion or in serum levels of these minerals were also found. When fed hemicellulose, the subjects were seen to be excreting significantly more zinc, magnesium, and copper than when fed cellulose or pectin.

Few studies have investigated hemicelluloses effects on divalent mineral balance in animals or humans. What research there is does indicate that hemicellulose does have the capacity to bind certain minerals and cause malabsorption to occur.

4. Oat Bran

Since the recent interest in the hypocholesterolemic effect of oat products in humans, several studies investigating the effect of oat products on mineral bioavailability have been published. As early as 1983, a study by van der Aar et al. (1983) used chicks as an animal model in determining divalent mineral utilization when these chicks consumed oat bran. They found that oat bran had no

significant influence on balance, excretion or serum mineral levels of calcium, magnesium, zinc, or copper in chicks.

In human studies, zinc absorption from meals based on rye, barley, oatmeal, triticale, or whole wheat showed a negative correlation between phytate and zinc absorption (Sandström et al., 1987). The dietary fiber with the lowest concentration of phytate (rye bread, 100 μmol phytic acid) had the highest absorption of ^{65}Zn (26.8 ± 7.4%) while the dietary fiber with the highest concentration of phytate (oatmeal porridge, 600 μmol phytic acid) had the lowest absorption of ^{65}Zn (8.4 ± 1.0%). These results must be interpreted with caution because the dietary fibers used were subjected to heat treatments prior to their ingestion. The authors stated that a wet heat treatment, such as boiling to prepare the porridge, had little effect on the phytic acid content of any of the cereals except for the rye flour. A reduction in the phytic acid content was seen when the cereals were subjected to a dry heat treatment as in bread baking. It was hypothesized that the heat treatment as such, changes physicochemical properties, e.g., of the fiber components, in a way that interferes with zinc absorption. The authors did not rule out that an incomplete isotope exchange may have occurred with the oat flakes

resulting in decreased labeled zinc being integrated in the raw oat flakes. They concluded that when phytate-rich cereals constitute a major portion of the total zinc intake of the diet, food preparation or other processes that reduce the phytic acid content can significantly improve the absorption of zinc.

The effects of oat and wheat-bran fibers on mineral metabolism in adult males was studied by Moak et al. (1987) in a 35 day metabolic diet study. For one week, all subjects consumed a vegetarian basal diet plus white bread. Following this control period, all subjects were assigned to either an oat-bran bread group or a wheat-bran bread group for 2 weeks, which was labeled the "High Bran" period. In a second period, oat and wheat-bran breads were reduced to half that of the high bran period which was labeled the "Low Bran" period and fed for 2 weeks. The authors failed to report how many slices of the wheat or oat-bran breads were consumed on a daily basis by the subjects for either periods. Diets and excretions (urine and fecal) were collected, composited, and analyzed for NDF and ADF fiber (food and feces) and for minerals (calcium, magnesium, zinc, and copper). Results indicated that both NDF and ADF fiber intakes were higher in the wheat-bran groups than in the oat-bran groups, regardless of the amount of added fiber.

Apparent balances of NDF in the oat-bran groups were higher than in the wheat-bran groups, while ADF balances were not different among any of the groups or periods. An estimation of hemicellulose content was not calculated by the investigators. Concerning minerals, it was determined that copper and zinc intakes were significantly higher during the High Bran period for both fibers as compared to the Low Bran period. Apparent copper balances were negative in all groups except the wheat-bran group of the High Bran period, which showed a positive balance. Fecal zinc excretion was highest in the oat-bran groups during both periods. Zinc balances were negative for the oat-bran groups and positive for the wheat-bran groups regardless of period. Calcium intakes and positive calcium balances were similar among both groups for both periods. Fecal excretion of calcium was significantly higher in the oat-bran group compared to the wheat-bran group during the High Bran period but not during the Low Bran period. Magnesium intakes were significantly higher for both groups during the High Bran period as compared to the Low Bran period. Negative apparent balances were found for both groups during both periods even though dietary intakes of magnesium were above the RDA level during the High Bran period and were 73.5% and 77.2% of the RDA in the oat-bran and wheat-bran groups,

respectively, during the Low Bran period. Total magnesium excretions were significantly greater in the oat-bran groups in both periods. Moak et al. (1987) concluded that oat-bran fiber appears to have a greater effect on zinc and magnesium metabolism than wheat-bran fiber; that oat-bran fiber has more effect on copper metabolism than wheat-bran fiber when fed in larger amounts; and that calcium intake or balance was unaffected by oat-bran fiber.

The effect of oat bran muffins on calcium absorption and calcium, phosphorus, magnesium,and zinc balance in men was investigated by Spencer et al. (1991). Eleven subjects consumed a basal diet for 40 days as an adaptation phase (baseline) prior to the start of the control period. During the experimental period the subjects consumed 4 oat bran muffins daily, two with breakfast and one each with lunch and dinner meals. Each muffin contained 21.8 g of oat bran and an average of 5.2 g of dietary fiber. The intake of the oat bran muffins increased the total dietary fiber intake from an average of 22.6 g/d in the control period to 43.2 g/d during the oat bran period. The calcium content of the diets was increased to RDA level by the supplementation with calcium gluconate tablets. Complete urine and stool collections were obtained throughout the control and experimental periods in addition to periodic blood samples.

Mineral analysis were performed on plasma/serum, diets, oat bran muffins and excreta but dietary fiber in the basal diet and oat bran muffins was calculated and not analyzed for. It was found that the consumption of the oat bran as muffins did not affect serum levels of calcium, phosphorus and magnesium or plasma level of zinc. Balance data indicated that urinary calcium excretion decreased significantly during the oat bran period, whereas fecal calcium excretion and calcium balance were unaffected. Calcium balances were negative for both the control and oat bran periods. Magnesium intake was significantly increased due to the consumption of the muffins. Both urinary and fecal magnesium excretions were increased significantly due to the muffin consumption but balance was not affected. Magnesium balance for the control period was negative while the balance during the oat bran period was positive. Zinc intake was slightly higher due to the consumption of the oat bran muffins but zinc balance was unchanged even though fecal zinc excretion was significantly increased in the oat bran period. Zinc balances for both the control and oat bran periods were positive. Spencer et al. (1991) concluded that the daily intake of 4 oat bran muffins containing a total of 21.8 g of oat bran did not have an adverse effect on calcium, magnesium or zinc balances from baseline

balances. The additional oat bran fiber used in this study had no effect per se on absorption or bioavailability of the minerals in question. In a recent study by Rossander-Hulthén, Gleerup and Hallberg (1990) oat bran and oat porridge were both found to inhibit non-haem iron absorption in human subjects by about 60% (study 1) and 45% (study 2). Radio-isotopes ^{59}Fe and ^{55}Fe were mixed into bread dough in order to trace iron absorption from the experimental meals. The test meals consisted of a continental-type breakfast with or without the addition of oat porridge (study 1) and a continental-type breakfast with or without extra oat fiber in the bread (study 2). Through analysis of phytate content and phytase activity the authors concluded that the absence of endogenous phytase activity was presumed due to the heat treatment used for commercial oat products to destroy lipase activity which prevents rancidity. This heat treatment possibly destroyed the phytase as well. They also found that the phytate in oats is more resistant to both endogenous and exogenous phytases in in-vitro experimentation with the oat bran. It was mentioned that the breakfast meal used did include other factors which affect iron absorption in man, such as coffee (inhibitory) and orange juice (enhances). The authors concluded that the

observed inhibition of iron by oat products is sufficiently high to be seriously considered if such products are regularly consumed.

From the research on oat bran's effects on mineral absorption in humans it appears that the consumption of oat bran does increase the dietary levels of zinc, magnesium and possibly that of calcium and copper. There is agreement that magnesium excretions, both urinary and fecal, appear to increase during oat bran consumption (Moak et al., 1987; Spencer et al., 1991). Fecal zinc excretion appears to increase due to oat bran consumption (Moak et al., 1987; Spencer et al., 1991). But contradictory effects are evident when looking at mineral balances. While Moak et al. (1987) found negative zinc, copper, and magnesium balances and a positive calcium balance; Spencer et al. (1991) found positive magnesium and zinc balances and a negative calcium balance with oat bran supplementation. Oat products have been reported to inhibit non-haem iron absorption in humans (Rossander-Hulthén et al., 1990).

E. Processing and Heat Treatments

Manufacturing and processing conditions at the commercial or home level may affect chemical composition of fiber which in turn may alter its physiological role in the

human body (Chang and Morris, 1990). In the processing of oats for human consumption, several steps occur that may alter the chemical composition or the physical structure of the oat grain. In processing of "green oats" to a form suitable for human consumption the grain is cleaned of foreign matter, screened and graded for adequate size (Burnette et al., 1992). The groats are then dried/roasted to decrease the moisture content using a gentle heating process. This "roasting" process serves several purposes such as:

- developing a fine flavor in the green-tasting or bitter groats
- improves the keeping qualities of the final product by inactivating lipases
- facilitates the removal of the hulls from the groats.

Although Burnette et al. (1992) refer to this roasting process as a "gentle heating", it is often for 1 to 2 hours at a temperature of 194°-212° F to inactivate lipases (Deane and Commers, 1986). At these temperatures and times, other enzymes could be also be affected, such as phytases. As part of their investigation, Rossander-Hulthén et al. (1990) investigated endogenous phytase activity in oats which had been heated to deactivate lipases. They found that dehusked oat groats, which had not been treated with any form of heat, showed a 23% reduction in phytate-P content during

incubation at 55° C at pH 5.5 for 24 hour. This is quite longer than the normal processing time for roasting. A similar incubation was preformed on oat bran but no change in phytate-P content from the original product was observed. This research indicates that roasting of "green" oat groats does have the possibility of altering components other than just the lipases. Caprez and Fairweather-Tait (1982) state that phytate is broken down by heat treatments and by phytase, an enzyme which occurs naturally in plant and also in the intestinal mucosa of man and rats. But, if phytase is inactivated by heating, can there be enough endogenous phytase to breakdown the phytate resulting in the release of minerals? Or is the effect directed more toward the heat treatment breaking down the phytate alone? In their study, Caprez and Fairweather-Tait (1982) incorporated into the diets of young male rats a bran based breakfast cereal (heated to 204° F for 40 minutes during manufacturing) at a level of 180g/kg of diet. A control group was fed an equal amount of bran that was not heat treated. They found that the phytate level of the untreated-bran diet (36.1 g/kg) was higher than the heat-treated-bran diet (23.9 g/kg). Phytase activity was not measured in this study. Zinc and iron availabilities were also investigated and it appeared that the heat treatment had no effect on zinc availability and

was not sufficient to improve zinc availability in the rat (Caprez and Fairweather-Tait, 1982).

After heating to drive off moisture, oat groats are usually milled further to physically change the grain to produce steel-cut oats, rolled flakes, quick and instant flakes, oat flour and oat bran (Burnette et al., 1992). Steel cutting converts the groats into uniform granules by cutting each groat into 2 to 4 pieces. These uniform pieces can then be steamed and rolled flat producing quick or instant oatmeal. Regular rolled oats are whole groats that are steamed and then rolled flat. The cutting and rolling increase surface area and reduce cooking time. In their study on heat treatment and particle size of bran on mineral absorption in rats, Caprez and Fairweather-Tait (1982) found that a reduction in particle size (like that seen with milling) resulted in slightly higher zinc retention in rats fed milled-bran diets as compared to rats fed course-bran diets.

Toasting of cereal products is performed to improve flavors and to impart color and crispness to the final product. Cooking that causes browning can increase the apparent fiber content of the food by the formation of artifact lignin via the Maillard reaction (Robertson, 1976). Lignin is known to bind minerals reducing availability.

Camire and Clydesdale (1981) investigated through in-vitro methods whether toasting, boiling and/or pH had any effects on the binding of calcium, magnesium, zinc, and iron to wheat bran, cellulose or lignin. Metal binding to fiber was found to be pH dependent. The toasting treatment had no effect on metal binding by cellulose, but had a significant effect on the binding of metals by lignin and wheat bran. Boiling had a significant effect on the binding of metals by all three fiber sources. The results of this study seem to lend support to the fact that lignin is very effective in binding metal ions and that the increase in some metals bound by wheat bran as a result of toasting may in fact be due to the small increase in the amount of "lignin" formed (Camire and Clydesdale, 1981).

To study the effect of heat processing treatments on insoluble dietary fiber (IDF), soluble dietary fiber (SDF), and total dietary fiber (TDF), Chang and Morris (1990) used in-vitro methods with oat bran, apple fiber, corn fiber, and soy fiber. The processing methods chosen were autoclaving at 121°C/15 minutes, at 100°C/30 minutes and microwave heating for 5 and 10 minutes. Chang and Morris (1990) found that microwave heating for 5 and 10 minutes significantly reduced the IDF and TDF of oat bran while no change in SDF was seen. Autoclaving resulted in significantly reduced TDF

content but showed no significant changes in IDF or SDF of oat bran. The authors stated that the effect of heat treatment appeared to depend on the fiber source and possibly on the relative quantities of IDF and SDF while different processing methods resulted in different effects on TDF. These results are in agreement with findings that cooking tends to decrease the soluble/insoluble fiber ratio of some foods (Mongeau et al., 1991; Shinnick et al., 1988). The alterations in the insoluble dietary fiber, soluble dietary fiber and total dietary fiber fractions could help explain mineral binding differences seen with processing treatments. Sandström et al. (1987) studied the zinc absorption in humans from meals based on rye, barley, oatmeal, triticale, and whole wheat. The oatmeal breakfast used as porridge was produced by wet heating (microwave heating for 12-20 minutes). While this method reduced the phytic acid content of rye flour it did not result in any significant changes in the phytic acid content of any of the other fiber sources studied. The authors suggest that the heat treatment used possibly changed the physiochemical properties of the fiber components (especially phytic acid) in a way that interfered with zinc absorption.

In the manufacture of some cereal products a process known as extrusion cooking is employed. Extrusion cooking

is generally a high temperature, short-time process using high shear at elevated pressure and is used to give texture to a wide variety of foods. Sandberg et al. (1986) found that extrusion cooking does appear to affect the digestibility of phytate. Kivistö et al. (1986) also report that extrusion cooking deactivates the phytase naturally present in wheat bran. They saw significantly decreased absorptions of zinc and magnesium but not of iron and calcium. Further studies by Sandberg et al. (1987) focused on the effect of extrusion cooking on phytate and mineral availabilities from wheat bran using ileostomy patients. They found that during extrusion cooking, 25% of the inositol hexaphosphate was degraded to penta- and tetraphosphates and that the phytase activity was lost. Essentially no phytate digestion occurred when the ileostomy patients consumed the extruded product. The digestibility of the inositol hexaphosphate of the extruded product was resistant to digestion in the human small intestine possibly due to loss of phytase activity or to the formation of the indigestible phytate complexes during the extrusion cooking (Sandberg et al., 1987). The undigested phytate in the ileal contents of the subjects resulted in decreased absorption of zinc and magnesium.

While most of the oat products intended for human

consumption are in the form of rolled oats and quick or instant oatmeals, some oats are further processed into flour and bran which can then be incorporated into a variety of food products. Flour can be used in bakery products, as thickeners and in ready-to-eat (RTE) cereals. Because of healthful claims for oat products, many commercial producers are trying to incorporate more of these products into the foods we use. Cereals, either hot cooked or RTE often use oats in one form or another. Morgan et al. (1986) examined breakfast consumption patterns using U.S.D.A. Nationwide Food Consumption Survey data and of the four most commonly consumed breakfasts, RTE cereal and milk was the second most often mentioned pattern. The authors found that when adults ate RTE cereal and milk their average daily intake levels of seven under-consumed nutrients increased. Also, on the average, those who ate cereal breakfasts had lower daily intakes of fat and cholesterol. This agrees with the idea that substitution of dietary fibers for foods that are rich in cholesterol and saturated fat help to decrease man's risk for cardiovascular disease (Sacks, 1991). Toma and Curtis (1989) point out that different RTE cereals contain varying amounts of dietary fiber, fat, sodium, simple sugars, and complex carbohydrates and that no "one" RTE cereal is perfect.

The effects of processing and heat treatments on dietary fiber appears to affect the chemical composition and physical form (Chang and Morris, 1990; Caprez and Fairweather-Tait, 1982; Sandström et al., 1987). Processing steps in oat production include heating to decrease the amount of lipases, which in turn may affect phytase levels (Rossander-Hulthén et al., 1990); toasting which may produce artifact lignin (Robertson, 1976); and cooking methods which appear to affect levels of soluble, insoluble and total dietary fiber (Chang and Morris, 1990). All of these factors have the potential of altering mineral binding by dietary fiber.

F. Tea, Coffee and Caffeine

Tea and coffee are two commonly consumed beverages by adult populations in most cultures. These beverages are made up of a complex array of compounds including polyphenols (sometimes called tannins), methylxanthines (especially caffeine), aluminum and other minerals, mainly magnesium and calcium (Greger and Lyle, 1988). Laboratory animal research has indicated that consumption of tea has adverse affects on zinc and iron absorption while improving absorption of copper in rats (Greger and Lyle, 1988; Fairweather-Tait et al., 1991).

Caffeine appears to affect calcium balance in rats. Yeh and Aloia (1988) fed rats a diet containing 4% instant coffee and a control group fed cellulose in place of the coffee. Three weeks of coffee consumption caused an increase in endogenous fecal calcium and urinary calcium excretion without a change in the absorption coefficient of calcium. Human studies involving coffee and/or caffeine on mineral availability are inconclusive. Massey and Hollingbery (1988) studied the acute effects of dietary caffeine on urinary mineral excretion in adolescents. They found that caffeine consumption resulted in the increased urinary excretion of calcium but not magnesium. They also showed that a sex/caffeine interaction was apparent with females exhibiting an increased urinary calcium concentration. Barger-Lux et al. (1990) showed that caffeine in doses of 400mg daily did not significantly effect calcium absorption, endogenous fecal or urinary calcium in premenopausal women. Beverages such as tea, coffee or other caffeine containing drinks may affect mineral availability in humans and need to be recognized as such.

MATERIALS AND METHODS

A. Materials[1]

Raw oat bran and processed ready-to-eat (RTE) oat bran were obtained from the Quaker Oats Company of Barrington, Illinois. Upon arrival the oat brans were stored at room temperature in the boxes in which they had been shipped. The raw oat bran (lot # 3LI5D) was in cardboard boxes containing 454 grams per box. The processed (RTE) oat bran (lot # 61, 81, and 83) was in cardboard boxes containing 426 grams per box.

The raw oat bran consisted entirely of "oat bran fraction". This fraction is obtained by passing the milled oat through a screen. The particles remaining on the screen correspond to the bran-rich fraction (oat bran), which is largely starch and protein, but contains a considerable amount of dietary fiber, of which about one-third is β-glucan gum (Quaker Oats Co., 1981).

The processed (RTE) oat bran contained the following ingredients: oat bran (75%), soy flour (12%), sugar (10%), and 3% minor ingredients (i.e., salt, calcium carbonate,

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[1] Taken from: Effects of Processing on Dietary Fiber Content of Oat Bran and Effects of Unprocessed and Processed Oat Bran on Plasma lipid levels of Healthy Males. Patricia Ann Ernest. (Thesis) VPI & SU, Dec. 1984.

sodium phosphate, L-lysine monohydrate, caramel color, niacinamide, FD & C Yellow No. 5, reduced iron, FD & C Yellow No. 6, BHA, riboflavin and thiamin monohydrate. An analysis of the raw and processed (RTE) oat brans is shown in Table 5.

TABLE 5

Composition of Raw and Processed (RTE) Oat Brans*

	Oat Bran[1]	
	RTE	Raw
Protein	20.3	19.7
Moisture	4.5	10.8
Fat	2.8	7.1
Ash	4.5	3.0
Fiber		
Crude (AOAC)	1.7	1.8
Total Dietary (proposed AOAC)	13.6	14.9

* From The Quaker Oats Company (1984)
[1] Values expressed as mean % of sample mass

B. Experimental Design

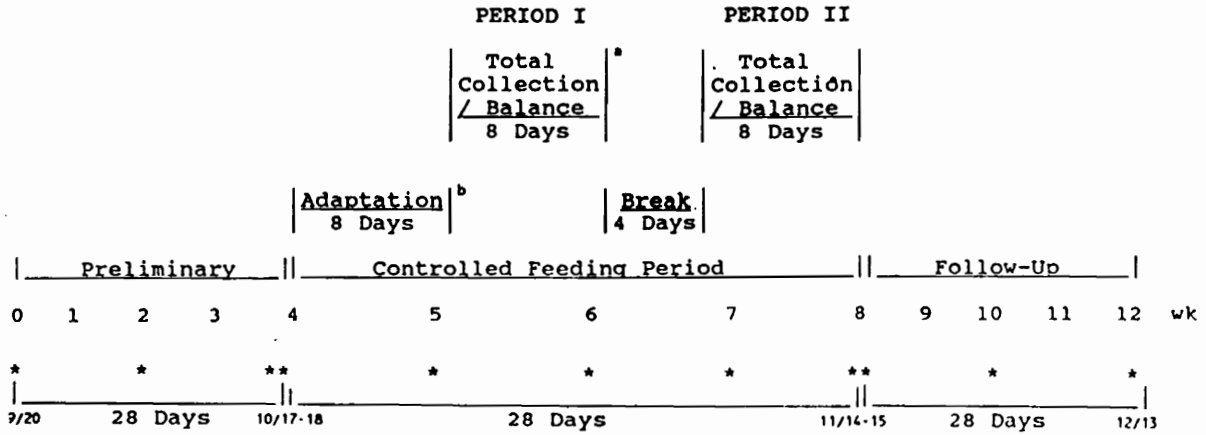
To study the effects of unprocessed and processed oat bran consumption on mineral balance, a metabolic diet study was undertaken in conjunction with an investigation studying the effects of unprocessed and processed oat bran on plasma lipids. Twenty-one male subjects were recruited from the faculty and graduate student populations at Virginia Polytechnic Institute and State University. To participate, the subjects had to have a fasting total cholesterol level of greater than 200 mg/dl as determined by a screening blood sample to accommodate the lipid part of the investigation. Each subject was confirmed, by personal physician or by University Health Service to be in good general health (Appendices A and B). All subjects were given both oral and written explanations of the study and written consent was obtained as required by the Institutional Review Board of Research with Human Subjects at Virginia Tech (Appendices C,D,E, and F).

The 21 subjects were assigned, using a random block design, to one of three dietary treatment groups; (1) control, no added fiber, (2) 100 g unprocessed oat bran added to the basal diet, (3) 100 g processed oat bran added to the basal diet. All diets were calculated to contain similar nutrient levels except for type and level of dietary

fiber. Each subject remained on the same dietary treatment for the duration of the experimental controlled feeding period which lasted 28 days. The diets were prepared, weighed, and served in the metabolic feeding unit on campus and subjects were instructed to consume only those food and drinks prepared at this unit. Occasionally, for the convenience of the subjects, meals or portions of meals were packaged and taken off the premises, but only with the approval of the investigators. Menu check sheets were provided at each meal and food consumption was monitored by the investigators (Appendix M). Any alterations were noted on the check sheets. Any consumption of food or drink not provided by the metabolic unit would result in dismissal from the study.

The study consisted of preliminary, controlled feeding, and follow-up periods of 4 weeks each (Figure 1 and Appendix G). A preliminary period was used as a baseline for usual nutrient dietary consumptions and blood mineral levels. Dietary intakes were determined from food recalls and 5-day dietary records kept by each subject.

Blood samples were drawn bi-weekly during the preliminary period, weekly during the controlled feeding and then bi-weekly during the follow-up period. The controlled feeding period was divided into seven 4-day periods using a



[a]October 26-November 3 (PERIOD I); November 7-15 (PERIOD II)

[b]October 18-26 (PERIOD I); November 4-7 (PERIOD II)

*Blood Sampling Dates

FIGURE 1

SCHEMATIC OUTLINE OF OAT BRAN METABOLIC FEEDING STUDY

4-day menu cycle. The first two menu cycles were used to adjust the subjects to the diet. Following this period of adjustment, an 8-day balance period was begun where all urine and fecal excretions were collected by each subject and used for mineral and fiber analysis. Another 4-day menu cycle was followed by a second 8-day balance period with urine and fecal collections. A 4-week follow-up period concluded the study with bi-weekly blood sampling.

During balance periods, the subjects were required to collect all urine and fecal excretions and were provided with appropriate containers. Plastic lined canvas tote bags were provided to each subject for carrying these containers. Blood sampling followed the outline in Appendix G. There was no attempt to collect sweat and subjects were asked not to alter their normal pre-study exercise levels. Subject weights were monitored daily throughout the experimental period so that any change in excess of 2 kg of the initial weight could be controlled (Appendix K). Addition of calories to maintain weight was given in the form of a food supplement (Table 6). Except for the dietary restrictions and collection of excreta, subjects were allowed to continue their normal activities.

TABLE 6

Composition of Supplement Unit

Food Item	Weight (g)
Processed Turkey	28
White Bread	56
Margarine	5

C. Experimental Diets

A basal diet was formulated to contain 4 g of dietary fiber with fat, protein, and carbohydrate making up 40, 12, and 48% of the calories, respectively. The intake of minerals was not brought to a given level, but was studied as it occurred with the dietary variations. The experimental diets were formulated from the basal diet by the addition of 100 g of a unprocessed oat bran[3] or 100 g of a processed ready-to-eat oat bran cereal[3] partially substituted for carbohydrate foods to provide an additional 15 to 16 g of dietary fiber. The control diet consisted of the basal diet plus carbohydrate foods low in dietary fiber. The diets consisted of conventional foods which were prepared under carefully controlled conditions to prevent possible mineral contamination. Distilled water was used for all drinking, cooking, and washing. To help eliminate mineral contamination, some disposable dinnerware, flatware, and cups were used. These disposable items were determined to be mineral free prior to their usage in the study.

Four different menus were fed on a cyclical basis throughout the study (Tables 7 through 11). Distilled water and coffee and tea prepared using distilled water were

[3]Quaker Oats Co., Barrington, IL.

TABLE 7

Food Items and Serving Sizes (in grams)
of the Menu for Day 1 for All 3 Experimental Diets

Food Item	Weight
Streusel Bar:	
butter	4.54
sugar	6.67
egg	2.28
pecans	4.72
white flour	5.48
Whole Milk	122.00
Pineapple Juice	250.00
Taco Spread:	
canned chickpeas	52.35
saltines	18.70
lemon juice	3.04
garlic powder	0.09
coriander	0.21
cumin	0.09
cayenne pepper	0.10
White Bread	56.00
Oleomargarine	14.20
Lettuce	14.00
Tomato	34.00
Cucumber	14.00
Apple Juice	248.00
Chocolate No-Bake Cookie	
cocoa	3.44
butter	9.07
whole milk	9.80
sugar	16.00
peanut butter	10.32

Food Item	Weight
Cheese Souffle:	
egg	32.00
whole milk	61.00
cayenne pepper	0.01
cheddar cheese	42.50
white bread	17.50
butter	5.88
Broccoli	99.00
Cream Sauce:	
oleomargarine	17.25
cornstarch	0.48
whole milk	37.04
lemon juice	5.56
marjoram	0.11
White Bread	56.00
Rice Pudding:	
brown rice, dry	20.00
vegetable oil	1.13
whole milk	115.46
heavy cream	30.00
egg yolk	8.50
brown sugar	13.75
vanilla extract	0.50
cinnamon	0.02
Raisins	20.68
Orange Juice	240.00
Saltines	28.00

TABLE 8

Food Items and Serving Sizes (in grams)
of the Menu for Day 2 for All 3 Experimental Diets

Food Item	Weight
Whole Milk	244.00
Grape Juice	95.00
Fresh Fruit Sauce:	
raisins	5.38
banana	43.75
orange sections	22.20
cottage cheese, 4% fat	56.75
Orange Juice	240.00
Tofu-Split Pea Spread:	
split peas, dry	17.71
onion	8.00
parmesan cheese	4.00
lemon juice	9.00
soy sauce	0.10
tofu	36.32
mayonnaise	2.24
Rice Pilaf:	
brown rice, dry	23.58
oleomargarine	4.02
whole wheat flour	1.41
whole milk	11.51
whole egg	5.17
lemon juice	2.00
basil	0.11

Food Item	Weight
Whole Wheat Bread	54.00
Butter	14.20
Corn Oil Margarine	12.50
Carrot	42.00
Apple, peeled	165.00
Parmesan Cheese	3.14
Nestea Ice Tea Mix with Lemon & Sugar	19.50
Tuna Salad:	
tuna, water packed	37.00
salad dressing	15.00
egg yolk, hard boiled	23.60
celery	15.00
coconut oil	8.40
olive oil	13.50
White Bread	56.00
"Parkay" Margarine	15.00
Potatoes, boiled no skins	109.00
Corn Oil Margarine	12.50
Chocolate Pudding	135.00
Cranberry Juice Cocktail	190.00

TABLE 9

Food Items and Serving Sizes (in grams)
of the Menu for Day 3 for All 3 Experimental Diets

Food Item	Weight
Peaches, canned	
with syrup	234.00
Banana	75.00
Whole Wheat Bread	54.00
Whole Milk	244.00
Butter	14.20
Eggplan Pate:	
eggplant, boiled and peeled	83.33
onion	5.00
egg yolk, hard boiled	11.80
garlic powder	0.09
parsley	0.40
olive oil	1.83
mayonnaise	28.00
White Bread	56.00
Butter	14.20
Cole Slaw:	
heavy cream	30.00
cabbage	70.00
sugar	6.00
cider vinegar	11.25
Pineapple Juice	250.00

Food Item	Weight
Ham Pizza:	
french bread	35.00
corn oil margarine	5.20
tomato sauce	65.50
mushrooms, canned	26.50
parsley	0.17
baked ham, 1% fat	28.00
mozzarella cheese	56.00
Tapioca Pudding:	
tapioca pudding mix	23.33
nutmeg	0.01
whole milk	119.60
Nestea Ice Tea Mix	
with Lemon & Sugar	19.50

TABLE 10

Food Items and Serving Sizes (in grams)
of the Menu for Day 4 for All 3 Experimental Diets

Food Item	Weight
Apple, peeled	165.00
Orange Juice	168.80
Whole Milk	244.00
Grape Juice	95.00
Chicken Salad:	
dark meat chicken, skinned, boiled	52.50
coconut oil	11.20
egg yolk, hard boiled	21.55
egg white, hard boiled	68.60
mayonnaise	28.00
celery	10.00
onion	10.00
parsley	0.10
Whole Wheat Bread	54.00
Vanilla Ice Cream	67.00
No-Bake Chocolate	
cocoa	3.44
butter	9.07
whole milk	9.80
sugar	16.00
peanut butter	10.32
Butter	10.00
Nestea Ice Tea Mix with Lemon & Sugar	19.50

Food Item	Weight
Chichpea Pate:	
chickpeas, canned	37.50
saltines	14.20
onion	7.50
garlic powder	0.09
parsley	0.50
lemon juice	5.00
cayenne pepper	0.01
black pepper	0.01
White Bread	28.00
Butter	10.70
Spaghetti, dry	55.42
Spaghetti Sauce:	
mushrooms, canned	100.60
tomato paste	28.00
tomatoes, canned, drained	38.00
canned tomato liquid	38.00
parsley	0.18
oregano	0.37
basil	0.37
salt	0.37
black pepper	0.37
Orange Juice	240.00

TABLE 11

Food Items & Serving Sizes (in grams)
Given to the 3 Experimental Diets on All 4 Menu Days

Food Item	Weight
Diet I:	
Cream of Wheat, dry	49.30
Sugar	7.60
Safflower Oil	3.70
Corn Oil Margarine	3.90
Orange Juice	100.00
Cottage Cheese, 1% fat	141.00
Diet II:	
Unprocessed "Mother's Oat Bran, dry	100.00
Sugar	3.00
Diet III:	
Processed (RTE) Oat Bran	100.00
Corn Oil Margarine	5.10
Safflower Oil	1.00

permitted with personal subject consumption noted on each subject's menu check sheet.

When needed, a food supplement was given to any subject whose weight was not maintained within 2 kg of initial weight. This supplement was in the form of a meat sandwich to supply 216 calories (Table 12).

D. Sample Collection

1. Food

a. Treatment Menus

At each meal, two extra portions of all food items were prepared for each treatment. Randomly, two were chosen from each treatment and the entire contents were placed into an appropriate 1-gallon paper carton (lined with two polyethylene bags) labeled with the group treatment and menu cycle day. This was done for each treatment for an entire day giving a total of six containers (two per each dietary treatment). At the end of the day, after all three meals had been added to the containers, each of the duplicate collections per treatment were weighed, recorded, and separately emptied into a tared 5-quart stainless steel commercial blender. All food was added and the plastic liners were rinsed several times with deionized water; this

TABLE 12

Food Items, Serving Sizes and Nutrient Analysis of the Supplement

Food Item	Serving Size (g)	Kcal	Pro (g)	CHO (g)	Fat (g)	SFA (g)	Lino* (g)	Fiber (g)
Processed Turkey	28	50	5	1	3	0.91	0.64	0.6
White Bread	56	130	5	28	2	0.40	0.40	0.0
"Parkay" Margarine	5	36	0	0	4	0.78	0.40	0.0
Total		216	10	29	9	2.09	1.44	0.6

*Linoleic Acid

was added to the blender container contents. Enough deionized water was added to ensure thorough blending. The blender container was then weighed and the weight was recorded. The food was homogenized for approximately 5 minutes at high speed and duplicate 5% aliquots were immediately taken and stored in appropriately labeled acid-washed, 1 liter plastic bottles. This process was repeated until food samples for an entire 4-day cycle had been added to the appropriate dietary treatment composite bottle. Composites were labeled and frozen at -4° C until analysis. Menu composites corresponding to the balance periods were the only ones analyzed. Supplements were not added to the menu composites.

b. Supplement

Supplement consumption differed for each subject due to weight variations. Consumption was recorded for each subject on a daily basis (Appendix O). Individual supplements were composited separately in the same manner as were the menus and each was analyzed similarly.

c. Coffee and Tea

Coffee and tea consumption for each subject differed due to individual preferences. Consumption was recorded for each subject on the menu check sheets and compiled into

daily totals (Appendix P). Samples of coffee and tea were analyzed on the basis of a single measure used to prepare the beverage. Only those minerals contributing one milligram or more were used in balance calculations.

2. Urine

During the balance periods the subjects were required to collect all of their urine in acid washed, 1-liter polyethylene bottles provided by the investigators. Collection bottles, graduated cylinders, and mixing jugs were labeled for each subject so that continual use of the same bottles by the same subject could be maintained. Acid washing consisted of using a 20% nitric acid solution to completely fill each bottle, graduated cylinder, and mixing jug and allowing the filled containers to sit for 30 minutes. Bottle caps were also acid washed in the same manner. The containers and caps were then rinsed thoroughly (6 times) with deionized water.

Urine collections were made on a 24-hour basis which began with the second voiding on day-1 and ended with the first voiding on day-2 throughout the balance periods. Each morning the subjects brought their urine to the metabolic lab for compositing as follows: urine bottles for each individual subject were emptied into 2-liter plastic,

acid washed graduated cylinders and the total volume was recorded. To thoroughly mix the urine, it was carefully poured into 1-gallon, acid washed plastic jugs and gently mixed. A 5% aliquot was taken and poured into labeled 1-liter, acid washed polyethylene bottles and frozen at -4° C for future mineral analysis. This procedure was repeated so that daily aliquots of urine were pooled for each subject for each of the 8-day balance periods.

The emptied urine collection bottles were then rinsed 3 times with both running tap water and deionized water. Before returning the bottles to the subjects, 2 ml of a 50% hydrochloric acid solution was added. The hydrochloric acid was added to prevent bacterial growth and possible nitrogen losses. Uric acid deposits were routinely removed from the bottles by using hot tap water, Alconox alkaline cleanser, and brushing with an acid washed brush. All of the other cylinders and jugs used were also rinsed three times with both running tap water and deionized water and inverted to air dry. Incomplete or missed collections were noted and resulted in that subject's data not being included in the statistical analysis.

3. Feces

All feces were collected in polyethylene bag lined 1-

quart, waxed cardboard containers labeled with the subjects name, subject number, date, and time of collection. These containers were previously determined to be free of mineral contamination. Fecal collections were made based on a counting method beginning the collections at 12 o'clock noon on day-2 of the collection period and continuing until 12 o'clock noon on the day after the feeding period ended. Incomplete or missed collections were noted and resulted in that subject's data not being included in the statistical analysis.

All fecal collections were frozen at -4° C until the end of the study at which time they were composited as follows: all feces from a given subject were partially thawed and separated on the basis of period number. The feces from a given period were then added to a tared 5-quart stainless steel commercial blender. All plastic liners of the fecal cups were carefully rinsed with deionized water and the water was added to the blender contents. The weight was recorded and the feces were homogenized for 5 minutes at high speed or until thoroughly blended. Labeled 500-ml and 250-ml acid washed polyethylene bottles were immediately filled with the homogenized fecal composite. The composites were refrozen at -20° C until mineral and fiber analysis could be performed. Care was taken to prevent extraneous

mineral contamination of the composites by the use of plastic gloves (when handling feces) and thorough rinsing of the blender cup between each composite with tap and deionized water.

4. Blood Plasma

In order to obtain blood samples, subjects were required to sign a blood donation consent form prior to the start of the study (Appendix N) in addition to the Consent of Participation - Nutrition Study form (Appendix E). Blood samples were taken on a routine basis throughout the study according to the schedule in Appendix G. Subjects were instructed to fast for at least 12 hours prior to blood sampling. Samples were drawn the morning after an overnight fast by approved medical personnel before the consumption of the breakfast meal. Multi-sample needles (21 gauge, 1½ inch) and trace mineral free vacutainers containing 143 units of sodium heparin as an anti-coagulant were used for all trace mineral blood collections. The labeled vacutainers were placed on ice when filled. Plasma was separated from red cells by centrifugation at 4° C for 30 minutes at 2000 x g. The plasma was pipetted from the packed red cells into polyethylene test tubes using plastic pipettes and stored -20° C until analysis. Both the plastic

pipettes and polyethylene test tubes were determined to be mineral free prior to their use.

D. Analytical Procedures

1. Food Analysis

At the end of the study, the frozen food composites and supplement composites were thawed and analyses were performed on the duplicate samples. Food aliquots were freeze-dryed by being frozen in plastic containers with a large surface area on the bottom. Once frozen, samples were placed in a freeze drier for 72 hours. Samples were then crushed by use of mortar and pestle to obtain a fine, homogeneous powder. Mortars and pestles were cleaned and dried between samples. Samples were then dried over night at approximately 50° C until no moisture could be removed. Samples were then stored in a desiccator until use. Total fat was extracted from freeze dried samples using petroleum ether (A.O.A.C., 1975). Nitrogen was determined on wet samples by using a modified Kjeldahl-Gunning-Arnold A.O.A.C. procedure (1975). Protein contents were then determined by multiplying the percent nitrogen by 6.25. Moisture was determined indirectly using the standard air oven method (A.O.A.C., 1975).

2. Dietary and Fecal Fiber

Fiber analysis of fecal and dietary composites were performed as described by Van Soest and Robertson (1977). Homogeneous fecal and food aliquots were frozen in plastic containers with a large surface area on the bottom. Once frozen, samples were placed in a freeze drier for 72 hours. Samples were then crushed by use of mortar and pestle to obtain a fine, homogeneous powder. Mortars and pestles were cleaned and dried between samples. Samples were then dried over night at approximately 50° C. Samples were then stored in a desiccator until analysis.

A modified neutral detergent fiber (NDF) method was employed using a heat stable α-amylase[4] (derived from Bacillus subtilis) as follows: ground, freeze-dried samples were further dried overnight at 50° C and then allowed to cool to room temperature in a desiccator. Approximately 0.5 g of the sample was weighed directly into 600 ml Berzelius beakers. All samples were analyzed in duplicate. To this, 50 ml of neutral detergent solution was added and the contents were swirled gently to mix. The beakers were brought to a boil and refluxed for 30 minutes with a condenser apparatus and heat. After 30 minutes, 50 ml of

[4]Sigma Chemical Company, St. Louis, MO.

refrigerated neutral detergent solution was added along with 2 ml of heat stable amylase to each beaker and the samples were allowed to reflux while boiling for another 30 minutes. The samples were then poured into weighed scintered glass crucibles to be filtered. All samples were rinsed from the beakers using hot, distilled water and samples in the crucibles were rinsed twice with hot, distilled water and twice with acetone. After filtering was complete, the Crucibles were dried overnight in a 100° C oven, cooled in a desiccator, weighed, and recorded. To account for ash content, the crucibles were then ashed in a muffle furnace at 500° C for 3 hours. The ashed crucibles were cooled, carefully removed from the muffle furnace to a desiccator, allowed to cool completely to room temperature, weighed, and the weight recorded. To calculate percent cell wall the following formula was used:

$$\frac{\text{crucible + dry sample weight} - \text{crucible + ashed sample weight}}{\text{initial sample weight}} \times 100 = \% \text{ cell wall NDF}$$

In order to estimate hemicellulose content, an acid detergent fiber (ADF) procedure was also performed on all food and fecal samples. This procedure was similar to the NDF method as follows: dried samples were weighed into Berzelius beakers and 100 ml of acid detergent solution were

added. The beakers were refluxed while boiling on a condenser apparatus with heating for 60 minutes. The samples were filtered into weighed scintered glass crucibles. All samples were rinsed from the beakers using hot distilled water and samples in the crucibles were rinsed twice with hot distilled water. Samples in crucibles were finally rinsed twice with hexane. Samples in the crucibles were dried overnight at 100° C, cooled in a desiccator to room temperature, weighed and recorded. As with the NDF samples, ash was accounted for by ashing crucibles in a muffle furnace at 500° C for 3 hours. The ashed crucibles were cooled, carefully removed from the muffle furnace to a desiccator and allowed to cool completely to room temperature, weighed and recorded. To calculate percent ADF the following formula was used:

$$\frac{\text{crucible + dry sample weight} - \text{crucible + ashed sample weight}}{\text{initial sample weight}} \times 100 = \% \text{ ADF}$$

To estimate hemicellulose, the ADF value for each sample was subtracted from its corresponding NDF value:

% NDF - % ADF = hemicellulose (estimated)

If duplicates differed by more than 5%, the NDF samples were rerun. Because of the small quantity of ADF present in

some samples the following cut-off points were imposed:

greater than 10% ADF -- 5% difference between duplicates allowed

less than 10% ADF -- 10% difference between duplicates allowed

Apparent neutral detergent fiber (NDF) and acid detergent fiber (ADF) digestibility were determined by the following formula:

$$\frac{\text{total fiber intake} - \text{total fiber output}}{\text{total fiber intake}} \times 100 = \% \text{ Apparent Digestibility}$$

Total fiber intake (either NDF or ADF) was obtained by taking the percent of fiber as determined by the NDF or ADF procedure for each of the treatment diets in the balance period and multiplying this amount by the amount of dry matter in the diet consumed during the balance period for each subject. To this was added the percent of NDF or ADF in each supplement, based on dry matter content, multiplied by the number of supplements eaten during each balance period by each subject. The total fiber output was obtained by taking the percent of fecal fiber, as determined by the NDF or ADF procedure, and multiplying this by the total dry matter fecal output for each subject during each balance period. These values were then put into the above formula

to determine apparent NDF or ADF digestibility.

3. Dietary and Fecal Minerals

Individual minerals analyzed for were calcium, magnesium, zinc, and copper using the wet ash (oxidation) procedure of Colin et al. (1983) as follows: duplicate 0.5 to 1.0 g samples of the thawed and thoroughly mixed composites were weighed (weights recorded) directly into labeled 50 ml acid washed beakers and covered with acid washed watchglasses. Mineral contamination was kept to a minimum by using separate plastic pipettes for sampling each composite. Two milliters of redistilled concentrated nitric acid and 1 ml of 60% reagent grade perchloric acid were added to each beaker, in that order. Caution was used when working with perchloric acid due to its reactive properties. The beakers covered with the watchglasses were allowed to digest on a hotplate in a specially designed fume hood for perchloric acid use. The temperature of the hotplate was initially 150° F, but then increased slowly to 325° F to allow samples to reflux until all organic matter had been digested. The beakers were watched closely at all times and temperatures were modulated to maintain a controlled reaction. Additional nitric acid was added when needed. Two blanks were run with each set of samples. When the

samples were completely digested and dense white fumes were generated, the watchglasses were lifted and the contents were allowed to dry under heat. The dried samples were then removed from the hotplate, cooled and the resulting ash rehydrated with 10 ml of 10% hydrochloric acid. The beakers were carefully covered with parafilm and allowed to sit for 24 hours so that the minerals would completely dissolve in the acid. Each beaker was poured into labeled polyethylene test tubes, stoppered, and stored under refrigeration until atomic absorption analysis.

Appropriate dilutions for magnesium (1:200), calcium (1:100), zinc (1:10), and copper (1:2) were then made and absorbencies were determined for each mineral using a Perkin-Elmer Model 503 Atomic Absorption Spectrophotometer[5] (AA) with acetylene as the fuel and air as the oxidant. To overcome phosphate and ionization interferences by sodium, calcium was diluted with a 0.3% lanthanum chloride solution. Mineral concentrations were determined by comparison of sample absorbencies to a standard absorbance curve using linear regression. Analyses were repeated if duplicates differed by more than 5%.

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[5]Perkin-Elmer Corp., Norwalk, CT.

4. Coffee and Tea Minerals

Samples of coffee and tea were analyzed on the basis of a single measure used to prepare the beverage. Minerals were quantified by diluting the samples with distilled, deionized water and aspirating directly into the AA. Mineral concentrations were determined by comparison of sample absorbencies to a standard absorbance curve using linear regression. Analyses were repeated if duplicates differed by more than 5%. Only those minerals contributing whole milligrams were used in balance calculations.

5. Urinary Minerals

Urine was used only for mineral analyses. The frozen composites were thawed and gently mixed. For zinc analysis, the urine was directly aspirated into the AA. For analysis of calcium and magnesium, the urine was appropriately diluted (Ca 1:80, Mg 1:200) with a 10% solution of hydrochloric acid and 0.3% lanthanum chloride and absorbencies were read on the AA. Copper content of urine was not measured because it has been shown in this laboratory to be negligible and of little significance in copper balance (Taper et al., 1980). Sample concentrations were calculated from appropriate standard curves using linear regression.

6. Plasma Minerals

Plasma calcium, magnesium, and zinc were analyzed for as follows: for zinc, plasma was diluted 1:5 with deionized water, with standards and blanks in a 5% glycerol solution to approximate the viscosity of the plasma. Calcium and magnesium were diluted 1:50 with a 0.3% lanthanum chloride solution with standards and blanks prepared from this same solution. Absorbencies were read from the AA and concentrations determined by comparison to standard curves using linear regression. Samples were rerun if duplicates differed by more than 5%.

E. Statistical Analysis

The computerized statistical analysis system (SAS)[6] was used to determine statistical significant differences. To test for a group*time interaction a multivariate analysis of variance (MANOVA) test was used. A significant effect was found for two of the study's data and therefore, the periods could not be combined.

An analysis of variance (ANOVA) model was used to determine significant differences among the three dietary treatments within the experimental periods with Duncan's

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[6]SAS Institute Inc., Cary, NC

Test (alpha = .05) to identify any effect of interaction between dietary treatment and period on mineral intake, urine and fecal excretion, and retention. Student's paired T-test ($P < 0.05$) was used to locate any significant differences within the same dietary treatments between the two experimental periods. All means are expressed as Least Square Means due to unequal group sizes. Absorption (mg/day), absorption (% of intake), Retention (mg/day), and retention (% of absorbed) were derived from measured data by the formulas:

Absorption (mg/day) = Intake - Fecal Excretion

$$\text{Absorption (\% of intake)} = \frac{\text{Intake - Fecal Excretion}}{\text{Intake}} \times 100$$

Retention (mg/day) = Intake - (Fecal + Urinary Excretion)

$$\text{Retention (\% of intake)} = \frac{\text{Intake - (Urine + Fecal excretion)}}{\text{Intake}} \times 100$$

Plasma mineral concentrations were tested for statistical significance first by fitting the plasma data to linear, quadratic, and cubic models. An alpha level of $P < 0.05$ was used to state statistical significance. Based on these models, further tests to determine if slopes were equal and if lines were parallel were employed. Analysis of variance (ANOVA) with contrasts was used to test for between

period (preliminary, controlled feeding, and follow-up) differences within the same treatment group and to test for significant differences between groups within the same periods.

RESULTS

A. Subject Information

Twenty-one males, between the ages of 20 and 60, were chosen to participate in the study. General physical characteristics of the individual subjects are listed in Table 13 with initial subject weights listed in Appendix J. All subject weights were considered to be normal. Most subjects lost weight during the course of the study. To increase caloric intakes a dietary supplement in the form of a meat sandwich (see Table 12) was given to keep the subject's weights to within 2 kg of their initial study weights. Subject's weights were monitored daily during the experimental period (Appendix K).

Two subjects, #8 and #18 dropped from the study early in the preliminary period. These two subjects are listed in Table 13 and Appendix J since their weights and total cholesterol levels were included in the random block design to determine the 3 treatment groups. Adherence to the study's protocol was strictly supervised by the investigators and determined to be excellent. However, during experimental period II, subject #9 became ill and reported incomplete urine and fecal collections. His data

TABLE 13

General Subject Information

SUBJECT #	AGE	HEIGHT[1]	WEIGHT[2]	WEIGHT[3]	BMI[4]	TC[5]
1	53	180.3	93.5	92.6	28.6	254
2	42	176.5	92.5	92.0	29.6	264
3	33	179.7	81.1	81.3	25.1	222
4	53	167.0	65.3	64.4	23.3	181
5	28	175.3	79.1	77.8	25.5	193
6	22	180.3	57.0	57.5	17.6	173
7	30	182.9	83.4	82.0	24.7	182
Mean			78.8	78.2	24.9	210
8 *	-	178.4	87.9	-	27.7	242
9	28	175.3	77.6	77.2	25.2	219
10	35	179.1	71.8	71.5	22.3	199
11	41	180.3	63.7	63.3	19.5	199
12	49	193.0	105.2	103.5	28.0	195
13	21	199.4	79.8	79.0	20.0	170
14	25	175.3	69.2	68.5	22.4	279
Mean			79.3	77.2	23.6	215
15	55	174.6	74.3	74.0	24.3	294
16	52	174.6	80.9	79.7	26.3	280
17	35	185.4	76.5	77.5	22.4	207
18 *	-	184.8	78.7	-	23.0	195
19	21	185.4	68.1	68.2	19.8	195
20	20	178.4	75.7	75.6	23.8	181
21	27	188.0	87.2	85.6	24.4	172
Mean			77.3	76.8	23.4	218

[1] In centimeters
[2] In kilograms, at start of experimental period
[3] At end of experimental period
[4] Body mass index, W/H^2 (kg/m^2), using mean of weight[2] and weight[3]
[5] Plasma total cholesterol at start of experimental period
* Subject who dropped from the study

was not included in the statistical analyses. Subject #3 sustained a fractured leg during period II and his data was also not used in the statistical analysis due to possible mineral alterations which occur with bone mending (Zeman,1990). This left six subjects in the control group and in the processed ready-to-eat (RTE) oat bran supplemented group and five subjects in the unprocessed oat bran supplemented group.

Medicine and/or drug usage was reported on the daily menu check sheets during the experimental period and are listed in Appendix L. Only one subject (#1) was routinely taking prescribed medication (6 to 9 aspirin per day). No significant usage was reported by any of the other subjects at any time during the course of the study.

B. Subject Intake Data

Individual subject data on the number of supplement units eaten per day and the number of cups (8 oz) of coffee, instant or brewed tea consumed per day are given in Appendices O and P. Intakes ranged from 0 to 7 for supplements, 0 to 8 for coffee and 0 to 4 for tea. The total number of supplement units consumed and cups of coffee and tea drunk per day per treatment group is shown in Table 14. Supplement unit intake was the highest for the

TABLE 14

Number of Supplement Units, and Amount of Coffee and Tea
Consumed per Day by Treatment Groups

GROUP		Day of Experimental Period																											
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28
CONTROL	Supplement	1	1	3	4	5	5	6	6	7	7	8	9	9	9	10	9	9	11	11	12	14	14	13	13	13	14	15	14
	Coffee	18	9	17	19	19	15	15	19	22	17	16	17	19	12	11	17	17	18	17	17	20	13	19	17	16	13	15	20
	Tea	2	1	0	1	1	0	1	1	1	0	1	2	4	4	1	1	3	2	2	3	3	4	4	3	5	4	3	3
UNPROC OAT BRAN	Supplement	6	7	12	11	12	13	14	14	13	14	13	13	14	13	14	13	13	12	11	13	14	15	14	14	13	15	14	14
	Coffee	7	2	6	6	9	6	7	6	7	7	7	5	8	7	4	6	7	5	7	7	7	4	6	5	7	7	7	5
	Tea	2	4	6	7	4	4	2	2	3	1	1	3	2	2	2	3	4	1	1	1	2	1	2	1	2	0	1	1
PROC OAT BRAN	Supplement	3	4	9	10	10	14	17	18	18	17	20	18	19	18	21	20	20	23	18	18	19	19	20	22	19	23	20	21
	Coffee	9	7	7	13	8	8	7	10	10	7	6	7	12	9	10	10	12	15	10	16	12	12	13	12	16	10	14	14
	Tea	3	4	4	5	6	4	2	3	5	2	1	3	0	0	2	1	1	1	1	1	2	0	0	1	0	0	0	0

unprocessed group (average of 17/day), intermediate for the processed RTE group (average of 13/day), and lowest for the control group (average of 9/day). The average daily intake of coffee was highest for the control group (2.4 cups/day) intermediate for the unprocessed group (1.8 cups/day) and lowest for the processed RTE group (1 cup/day). The average daily intake of tea was approximately the same for each group.

C. Dietary Information

1. Menu

The analysis of each diet for the two controlled feeding collection periods is presented in Table 15. All data reported are from actual nutrient analysis performed at the Department of Human Nutrition and Foods at Virginia Tech. Each experimental collection period was made up of two cycles of a 4-day menu plan for a total period length of 8 days each. The grams of protein contained in the three diets were nearly equal with only slight variations among the three groups for both periods. Dietary fat content of the oat bran diet was highest, the RTE intermediate and the control diet lowest for both periods. Moisture content of each diet was similar for both periods. Moisture content of the control diet was highest with both experimental diets

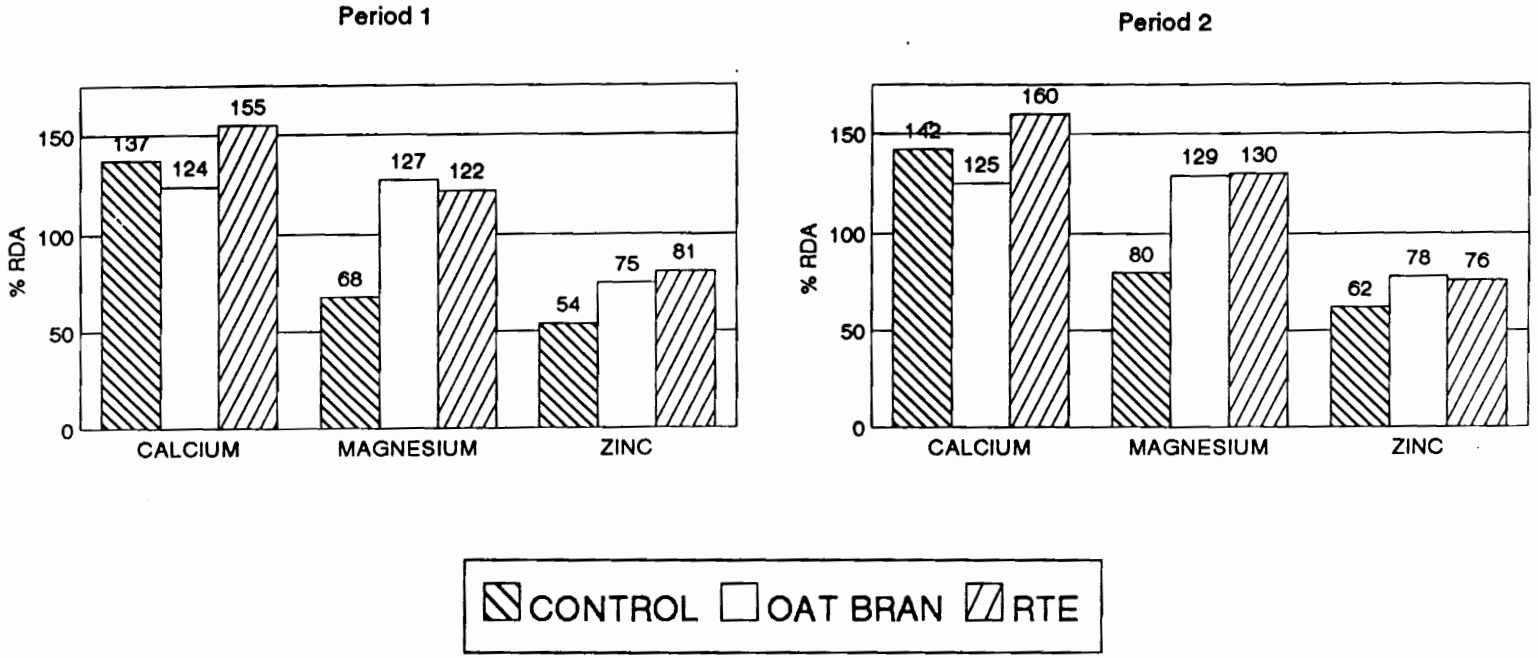
TABLE 15

Protein, Fat, and Moisture Content of Treatment Diets

Diet	Grams Protein Per Day	Grams Fat Per Day	Percent Water
Period I			
Control	83	105	80.5
Oat Bran	83	130	77.1
RTE	81	112	77.9
Period II			
Control	85	104	80.7
Oat Bran	84	120	78.1
RTE	83	111	72.4

containing slightly less. Mineral content of the diets expressed as percent of the RDA for period I and II are presented in Figures 2 and 3. Mineral content of the diets remained relatively constant over the two experimental periods. Calcium was above the current RDA recommended level of 800 mg/day for all treatment groups during both periods with only slight variations between periods. The inclusion of oat bran appeared to increase dietary magnesium to levels above the current RDA recommended level of 350 mg/day for adult males during both periods. The control group diet contained less than the RDA recommended level for magnesium. Likewise, the zinc content of the diets also appeared to be increased when oat bran was added, but was below the RDA recommended level of 15 mg/day. There does not appear to be any difference that can be attributed solely to processing treatment of the oat bran for these differences in mineral content. Dietary copper content was within the ESADDI range of 1.5 - 3.0 mg/day for all three groups during both periods. The inclusion of oat bran, regardless of processing treatment, does appear to have improved the copper content of the experimental diets with both the fiber supplemented groups nearly equal in their dietary copper content.

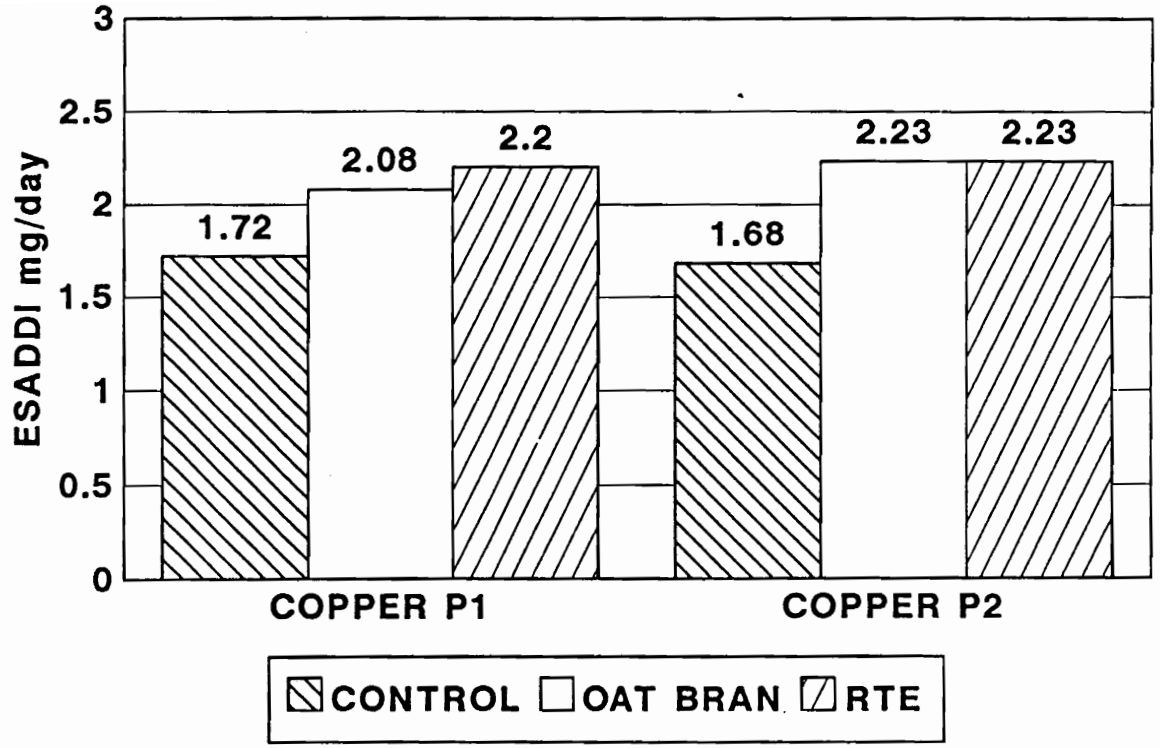
Dietary fiber contents (NDF, ADF, and estimated



RDA--Recommended Dietary Allowances (NRC, 1989)

FIGURE 2

MINERAL CONTENT OF EXPERIMENTAL DIETS AS PERCENT OF RDA



ESADDI--Estimated Safe and Adequate Daily Dietary Intakes (NRC, 1989)

FIGURE 3
COPPER CONTENT OF EXPERIMENTAL DIETS COMPARED WITH ESADDI
Period 1 and Period 2

hemicellulose) of the diets are presented in Table 16. During Period I, the addition of oat bran appeared to cause an increase in the dietary NDF present for both experimental groups. During Period II only the unprocessed Oat Bran diet contained a higher percentage of NDF fiber than the two other diets. Percent ADF fiber was relatively constant for all groups during both periods with Period II showing a slightly higher percentage. The percent calculated hemicellulose closely followed the variations seen for percent NDF fiber since this measure is dependent upon percent NDF.

2. Supplement

Moisture, fat, protein, fiber, and mineral contents of the supplement are presented in Table 17. The supplement unit was designed to increase the caloric intake without increasing the fiber intake. Analysis indicated that the supplement was high in fat and relatively low in protein. Fiber analysis showed that the supplement contained nearly equal amounts of NDF and ADF with only minimal estimated hemicellulose. The mineral content of the supplement followed the order of:

zinc < copper < calcium < magnesium

In determining actual mineral and dietary fiber

TABLE 16

OAT BRAN STUDY
MENU DIETARY FIBER CONTENT

Group	Period	% NDF	% ADF	Estimated Hemicellulose
Control	1	6.79	3.17	3.62
Oat Bran	1	8.46	3.88	4.58
RTE	1	7.22	3.46	3.76
Control	2	6.40	4.23	2.17
Oat Bran	2	7.56	4.07	3.49
RTE	2	6.90	4.27	2.63

NDF = neutral detergent fiber
ADF = acid detergent fiber
Estimated Hemicellulose = NDF - ADF

TABLE 17

Protein, Fat, and Moisture Content of Dietary Supplement Used in Oat Bran Study

% H_2O	% Fat	% Protein
93.7%	25.5%	3.29%

Oat Bran Study Supplement
Dietary Fiber Content

% NDF	% ADF	Estimated Hemicellulose
2.46%	2.40%	0.06%

Mineral Content of Supplement Consumed During Oat Bran Study

	mg/each supplement
Calcium	0.32
Magnesium	0.45
Zinc	0.01
Copper	0.06

intakes, the number of supplements eaten per controlled collection period for each subject was added to the menu dietary intake (see Appendix O).

3. Beverages

Mineral analyses of the beverages consumed are presented in Table 18 and reported as mg/serving. Only those values contributing one milligram or more per serving were included in the total mineral intake for each subject. Calcium from coffee and magnesium from all beverages were high enough for inclusion in calculating total dietary intake. Zinc and copper contents of the beverages were minimal and were not included in calculating total dietary intake. Beverage consumption was recorded on the daily menu check sheets at each meal (Appendix P). These beverages were consumed only with the meals; any other time unlimited distilled water was available. Mineral intakes from the beverages were added to the dietary and supplement mineral intakes to obtain total mineral intakes for each subject.

D. NDF, ADF, and Calculated Hemicellulose Content of Diets

Mean intakes of NDF, ADF, and the calculated hemicellulose content of the diets consumed by the three experimental groups are shown in Table 19 with individual

TABLE 18

MINERAL CONTENT OF THE BEVERAGES CONSUMED DURING OAT BRAN STUDY*

	Ca	Mg	Zn	Cu
Instant Tea**	0.50	5.81	0.20	0.01
Brewed Tea***	0.40	5.82	0.11	0.02
Coffee+	9.12	5.92	0.17	0.00

*reported as mg/ea. serving -- only those values underlined are included in the total mineral intake

**based on an average serving size of 0.60 g reconstituted with with distilled water

***based on one tea bag steeped for 3 minutes in hot distilled water

+based on an average serving size of 1.65 g reconstituted with hot distilled water

Table 19

Mean Intake of NDF and ADF Fiber and Calculated Hemicellulose of Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Total NDF Intake g/day	36.9^{a}	50.2^{c1}	41.1^{b1}	36.5^{a}	43.7^{c2}	39.8^{b2}
SEM	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3
Total ADF Intake g/day	17.5^{a1}	23.6^{c}	20.3^{b1}	24.4^{ab2}	24.0^{a}	25.1^{b2}
SEM	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3
Est. Hemi-Cellulose** g/day	19.4^{a1}	26.7^{c1}	20.8^{b1}	12.1^{a2}	19.7^{c2}	14.7^{b2}
SEM	± 0.01	± 0.01	± 0.01	± 0.01	± 0.01	± 0.01

**Estimated hemicellulose = %NDF minus %ADF

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

values listed in Appendix Q. The addition of oat fiber resulted in increased ($P < 0.05$) NDF intakes for the fiber supplemented groups. The unprocessed oat bran group consumed the highest amount of NDF during both experimental periods, the processed RTE group consumed intermediate amounts and the control group consumed the lowest amount. Significant differences were seen within the same dietary treatments between periods for the fiber supplemented groups, with Period I NDF consumptions higher than Period II ($p < 0.05$). The control group's NDF intake remained constant over both periods.

Total ADF intakes were significantly increased for the fiber supplemented groups ($P < 0.05$). The oat bran group consumed the highest amount, the RTE group intermediate, and the control the lowest during Period I. During Period II, the oat bran group was significantly lower than the RTE group ($P < 0.05$). Significant differences were seen within the same dietary treatments between periods for the control and the RTE groups with Period II ADF intakes higher than Period I ($P < 0.05$). The unprocessed oat bran group's consumption of ADF was similiar for both experimental periods.

The estimated hemicellulose consumptions largely reflected the NDF intakes. The oat bran group consumed the

most, the RTE group intermediate, and the control group the least amount of estimated hemicellulose during both experimental periods ($P < 0.05$). Significant differences were seen within the same dietary treatments between periods with Period I consumptions higher than Period II ($P < 0.05$).

E. Digestibility

1. NDF Intake, Output, and Digestibility

Mean NDF intake, output, and percent digestibility for both experimental periods are shown in Table 20 with individual data listed in Appendix R.

Significant differences in total NDF intake were seen during both periods. The unprocessed oat bran group had the highest NDF intakes, the processed RTE group intermediate and the control group the lowest intakes during both periods ($P < 0.05$). Between periods, both the oat bran and RTE groups showed a significantly higher NDF intake during Period I than during Period II ($P < 0.05$). Total NDF intakes for the control group were similiar during both experimental periods.

No significant differences were seen for total NDF output for any of the groups during Period I. During Period II the oat bran group excreted significantly more NDF than the control group ($P < 0.05$).

Table 20

Mean Neutral Detergent Fiber Intake, Output, and Digestibility of Diets by Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Total NDF Intake g/day	36.9[a]	50.2[c1]	41.1[b1]	36.5[a]	43.7[c2]	39.8[b2]
SEM	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3
Total NDF Output g/day	6.1	10.4	8.6	5.3[a]	12.3[b]	8.8[ab]
SEM	± 1.7	± 1.8	± 1.7	± 1.7	± 1.9	± 1.7
Percent NDF Digestibility	83.6%	79.3%	79.2%	85.5%[b]	71.9%[a]	77.9%[ab]
SEM	± 3.5	± 3.9	± 3.5	± 4.0	± 4.4	± 4.0

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

Percent NDF digestibilities during Period I were similiar for all three experimental groups. Oat fiber supplementation was seen to depress the digestibilities. During Period II, the oat bran group NDF digestibility was significantly less than the control group ($P < 0.05$).

The addition of oat fiber, especially unprocessed, does appear to increase NDF intakes and resulted in decreased NDF digestibility during Period II.

2. ADF Intake, Output, and Digestibility

Mean ADF intake, output, and percent digestibility for both experimental periods are shown in Table 21 with individual data listed in Appendix S.

Significant differences in total ADF intake were seen during both periods. The unprocessed oat bran group had the highest ADF intake, the RTE group intermediate and the control group the lowest during Period I ($P < 0.05$). During Period II, the processed RTE group's ADF consumption was significantly higher than the oat bran group ($P < 0.05$). Between periods, both the control and RTE groups showed significantly higher ADF intakes during Period II than during Period I ($P < 0.05$). Total ADF intakes for the oat bran group were similiar during both experimental periods.

ADF output of the oat bran group was significantly

Table 21

Mean Acid Detergent Fiber Intake, Output, and Digestibility of Diets by Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Total ADF Intake g/day	17.5[a1]	23.6[c]	20.3[b1]	24.4[ab2]	24.0[a]	25.1[b2]
SEM	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3	± 0.3
Total ADF Output g/day	1.7[a]	5.7[b]	4.3[ab]	1.9[a]	6.3[b]	4.3[ab]
SEM	± 1.0	± 1.1	± 1.0	± 1.0	± 1.1	± 1.0
Percent ADF Digestibility	90.2%[b]	75.7%[a]	79.3%[ab]	92.1%[b]	73.9%[a]	83.1%[ab]
SEM	± 4.4	± 4.8	± 4.4	± 4.2	± 4.6	± 4.2

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

higher than the control group during both experimental periods ($P < 0.05$). ADF outputs for the RTE group were similiar during both periods.

The control group had significantly higher ADF percent digestibilities than the unprocessed oat bran group during both experimental periods ($P < 0.05$).

Oat fiber supplementation, regardless of processing treatment was seen to increase ADF intake during Period I. During Period II, the processed fiber supplemented group appeared to have increased ADF intake above that of the unprocessed group. Unprocessed oat fiber supplementation appeared to have increased ADF output and accordlingly decreased ADF digestibility.

F. Calcium

Calcium Intake

Mean calcium intake, excretion, retention and percent apparent retention are shown in Table 22 and individual data are shown in Appendix T. Mean calcium intakes varied from the diet content due to the individual additions of supplement units and coffee consumed per day. Consistently across both experimental periods the groups consumed calcium in the following order:

Oat Bran < Control < RTE

Table 22

Mean Calcium Intake, Excretions, Absorption, and Retention in Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Intake						
mg/day	1098[b1]	993[a1]	1137[c1]	1137[b2]	1004[a2]	1281[c2]
± SEM	±0.2	±0.3	±0.2	±0.3	±0.3	±0.3
Excretion						
Urine	235	145	207	243	183	214
± SEM	± 33	± 36	± 33	± 29	± 32	± 29
Feces	785[a]	818[a]	985[b1]	880[a]	846[a]	1156[b2]
± SEM	± 47	± 52	± 47	± 48	± 73	± 48
Absorption						
mg/day	313[a]	175[ab]	152[b]	257	158	125
± SEM	± 47	± 51	± 47	± 48	± 53	± 48
% of Intake	28.5%	17.6%	13.3%	22.6%	15.8%	9.8%
± SEM	± 4	± 5	± 4	± 4	± 5	± 4
Retention						
mg/day	78	30	-55	14	-25	-89
± SEM	± 70	± 76	± 70	± 66	± 73	± 66
% of Intake	7.1%	3.0%	-4.8%	1.2%	-2.4%	-6.9%
± SEM	± 6	± 7	± 6	± 6	± 6	± 6

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

with each group significantly different from each other ($P < 0.05$). Between Period I and Period II, significant differences were seen within the dietary treatments with Period II data significantly higher than Period I ($P < 0.05$).

Calcium Excretion

Mean daily urinary calcium excretions ranged from 145 ± 36 to 243 ± 32 mg for both periods. No significant differences were seen between periods, within treatments, or among treatment groups for urinary calcium excretion.

Mean daily fecal calcium excretions ranged from 785 ± 47 to 1156 ± 66 mg for both periods. Results showed a significant difference in fecal calcium excretion with the RTE group excreting significantly more fecal calcium than either the control or oat bran groups ($P < 0.05$). Between periods, the RTE group showed a significant increase in fecal calcium excretion from Period I to Period II ($P < 0.05$).

Calcium Retention

Mean daily calcium retention values ranged from -89 ± 66 to 78 ± 70 mg for both periods with individual data ranging from -352 to 294 mg/day. Nine of the 17 subjects

were in negative balance during Period I. Ten subjects were in negative balance during Period II. No significant differences in apparent calcium retention were seen between periods, within treatments, or among treatment groups. During Period I the RTE group was in negative balance while during Period II both of the fiber supplemented groups were in negative balance.

G. Magnesium

Magnesium Intake

Mean magnesium intake, excretion, retention, and apparent absorption are shown in Table 23 with individual data shown in Appendix U. Mean magnesium intakes varied from the diet content due to the individual additions of supplement units, coffee, and tea consumed per day. Significant differences were seen within the same dietary treatments between periods with Period II magnesium intakes higher than Period I ($P < 0.05$). The experimental groups were seen to be significantly different ($P < 0.05$) from each other during Period I in the following order:

Control < RTE < Oat Bran

For Period II the groups were still significantly different ($P < 0.05$) from each other but the order changed to:

Control < Oat Bran < RTE

Table 23

Mean Magnesium Intake, Excretions, Absorption, and Retention in Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Intake						
mg/day	239.4[a1]	444.8[c1]	428.0[b1]	281.0[a2]	450.2[b2]	456.0[c2]
± SEM	± .04	± .04	± .04	± .04	± .04	± .04
Excretion						
Urine	87.5	68.7	98.3	99.8	85.6	109.1
± SEM	± 14	± 15	± 14	± 13	± 14	± 13
Feces	151.7[a]	306.3[b]	280.2[b]	164.3[a]	304.1[b]	337.4[b]
± SEM	± 47	± 52	± 47	± 59	± 65	± 59
Absorption						
mg/day	88[a]	138[b]	148[b]	117	146	119
± SEM	± 15	± 17	± 15	± 21	± 23	± 21
% of Intake	36.6%	31.1%	34.5%	41.5%	32.5%	26.0%
± SEM	± 4	± 5	± 4	± 5	± 6	± 5
Retention						
mg/day	0.2[a]	69.8[b]	49.5[b]	17.0	60.6	9.5
± SEM	± 18	± 19	± 18	± 21	± 23	± 21
% of Intake	0.1%	15.7%	11.6%	6.0%	13.5%	2.1%
± SEM	± 5	± 6	± 5	± 5	± 6	± 5

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

Magnesium Excretion

Mean daily urinary magnesium excretions ranged from 68.7 ± 15 to 109.1 ± 13 mg for both periods. No significant differences were seen between periods, within treatments, or among treatment groups for urinary magnesium excretion.

Mean daily fecal magnesium excretions ranged from 151.7 ± 47 to 337.4 ± 59 mg for both periods. Results showed significant differences in fecal magnesium excretion during both experimental periods with the fiber supplemented groups excreting significantly more fecal magnesium than the control group ($P < 0.05$).

Magnesium Retention

Mean daily magnesium retention data ranged from 0.2 ± 18 to 69.8 ± 19 mg for both periods with individual data ranging from -103.3 to 128.7 mg/day. In Period I three subjects were in negative magnesium balance (two in control group and one in RTE group); while in Period II, five subjects were in negative magnesium balance (two in the control group, one in the oat bran group, and two in the RTE group). The mean group retentions were positive of all three groups for both periods. During Period I, the oat bran group retained significantly more magnesium than the control group ($P < 0.05$). During Period II there were no

significant differences among the treatment groups.

H. Zinc

Zinc Intake

Mean zinc intake, excretion, retention, and apparent absorption are shown in Table 24 with individual data shown in Appendix V. Mean zinc intakes varied from the diet content due to the individual additions of supplement units consumed per day. Significant differences were seen within the same dietary treatments between periods with Period II zinc intakes higher than Period I for the control and oat bran groups ($P < 0.05$); but opposite for the RTE group, with Period II significantly higher than Period I ($P < 0.05$). The experimental groups were seen to be significantly different ($P < 0.05$) from each other in the following order during Period I:

Control < Oat Bran < RTE

For Period II the groups were still significantly different ($P < 0.05$) from each other but the order changed to:

Control < RTE < Oat Bran

Zinc Excretion

Mean daily urinary zinc excretions ranged from 0.58 ± 0.36 to 0.85 ± 0.13 mg for both periods. No significant

Table 24

Mean Zinc Intake, Excretions, Absorption, and Retention in Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Intake						
mg/day	8.05[a1]	11.20[b1]	12.10[c1]	9.30[a2]	11.67[c2]	11.34[b2]
± SEM	±0.0	±0.0	±0.0	±0.0	±0.0	±0.0
Excretion						
Urine	0.78	0.58	0.81	0.71	0.70	0.85
± SEM	± .13	± .14	± .13	± .13	± .14	± .13
Feces	7.39[a]	9.65[b]	9.30[b]	7.48[a]	9.61[b]	9.75[b]
± SEM	± .58	± .64	± .58	± .59	± .65	± .59
Absorption						
mg/day	0.66[a]	1.56[ab]	2.80[b]	1.83	2.06	1.59
± SEM	± .6	± .6	± .6	± .6	± .6	± .6
% of Intake	8.2%	13.9%	23.2%	19.7%	17.6%	14.0%
± SEM	± 6	± 6	± 6	± 5	± 6	± 5
Retention						
mg/day	-0.12[a]	0.98[ab]	1.99[b]	1.12	1.35	0.74
± SEM	± .6	± .6	± .6	± .6	± .6	± .6
% of Intake	-1.44%	8.74%	16.47%	11.99%	11.61%	6.55%
± SEM	± 6	± 7	± 6	± 6	± 6	± 6

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

differences were seen between periods, within treatments, or among treatment groups for urinary zinc excretion.

Mean daily fecal zinc excretions ranged from 7.39 ± 0.58 to 9.75 ± 0.59 mg for both periods. Results showed significant differences in fecal zinc excretion during both experimental periods with the fiber supplemented groups excreting significantly more fecal zinc than the control group ($P < 0.05$).

Zinc Retention

Mean daily zinc retention data ranged from -0.12 ± 0.59 to 1.99 ± 0.59 mg with individual data ranging from -1.90 to 4.36 mg/day. No significant differences were seen between periods, within treatments, or among treatment groups for zinc retention. The control group in Period I was the only group, regardless of experimental period, in negative balance.

I. Copper

Copper Intake

Mean copper intake, excretion, retention, and apparent absorption are shown in Table 25 with individual data shown in Appendix W. Mean copper intakes varied from the diet content due to the individual additions of supplement units

Table 25

Mean Copper Intake, Excretion, Absorption, and Retention in Subjects Consuming Two Types of Oat Bran

	Period I			Period II		
	Control	Oat Bran	RTE	Control	Oat Bran	RTE
Intake						
mg/day	1.72[a1]	2.08[b1]	2.20[c1]	1.68[a2]	2.23[b2]	2.23[b2]
± SEM	±0.0	±0.0	±0.0	±0.0	±0.0	±0.0
Excretion						
Feces	1.38	1.68	1.72	1.31[a]	1.43[ab]	1.80[b]
± SEM	± .11	± .13	± .11	± .14	± .15	± .14
Absorption						
mg/day	0.34	0.40	0.48	0.37	0.80	0.43
± SEM	± .12	± .13	± .12	± .14	± .15	± .14
% of Intake	19.6%	19.4%	21.7%	22.0%	35.7%	19.4%
± SEM	± 6	± 7	± 6	± 7	± 7	± 7
Retention						
mg/day	0.34	0.40	0.48	0.37	0.80	0.43
± SEM	± .12	± .13	± .12	± .14	± .15	± .14
% of Intake	19.6%	19.4%	21.7%	22.0%	35.7%	19.4%
± SEM	± 6	± 7	± 6	± 7	± 7	± 7

[a] Values with different superscripts within periods are significantly different ($P < 0.05$).

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.05$).

consumed per day. Significant differences were seen within the same dietary treatments between periods. Period I intake for the control group was lower than Period II ($P < 0.05$). Both of the fiber supplemented groups consumed less copper during Period I than in Period II ($p < 0.05$). The addition of oat bran, regardless of processing treatment, increased the copper content of the experimental diet. All three groups were consuming an adequate level of copper as compared to the ESADDI.

Copper Excretion

Mean daily fecal copper excretions ranged from 1.31 ± 0.14 to 1.80 ± 0.14 mg. No significant differences were seen during Period I, but, results showed that during Period II the RTE group excreted significantly more fecal copper than the control group ($P < 0.05$).

Copper Retention

Mean daily copper retention values ranged from 0.33 ± 0.12 to 0.80 ± 0.15 mg with individual data ranging from -0.58 to 1.02 mg/day. During Period I only two subjects were in negative copper balance (subject #2 in the control group and subject #11 in the oat bran group). During Period II, only one subject was in negative balance

(subject #20 in the RTE group). Mean copper retention for all experimental groups during both periods were positive. No significant differences were seen between periods, within treatments, or among treatment groups for copper retention.

J. Plasma Mineral Concentrations

Calcium

Mean weekly plasma calcium data are shown in Table 26 with individual data listed in Appendix X. Graphic representation of mean plasma calcium over elapsed time of the study in days is shown in Figure 4. Mean weekly plasma calcium data ranged from 9.05 ± 3.3 to 10.47 ± 2.9 mg/dl with individual data ranging from 8.58 to 10.99 mg/dl. When fitted to linear, quadratic and cubic models plasma calcium failed to show any linear, quadratic or cubic effects. After testing for model fit the mean plasma calcium data for each division of the study (preliminary, controlled feeding and follow-up) were pooled and ANOVA statistics performed on the pooled means.

During the preliminary period, plasma calcium data for all three treatment groups were significantly higher than the plasma data during the controlled feeding or follow up periods ($P < 0.002$). During the preliminary period a significant between group difference was found with the oat

Table 26

Mean Plasma Calcium Values of Subjects Consuming Two Types of Oat Bran*

	Period										
	Preliminary				Controlled Feeding					Follow Up	
Group	9/20	10/04	10/17	10/18	10/25	11/01	11/08	11/14	11/15	11/29	12/13
Control	9.67	10.19	10.32	10.04	9.21	9.26	9.47	9.58	9.22	9.29	9.57
± SD	1.8	3.0	2.5	3.2	2.6	3.0	2.1	3.9	3.3	5.0	4.1
			10.11^{a1}				9.48^{a2}				9.39^{2}
Oat Bran	9.42	9.94	10.05	9.68	9.23	9.15	9.53	8.97	9.05	9.38	9.27
± SD	3.2	6.2	3.9	4.3	3.4	4.3	2.1	2.5	3.3	5.0	2.4
			9.84^{b1}				9.37^{b2}				9.23^{2}
RTE	9.72	10.08	10.47	10.02	9.35	9.62	9.56	9.68	9.42	9.45	9.82
± SD	0.8	5.7	2.9	3.2	2.0	1.9	3.0	2.9	0.8	2.0	4.1
			10.14^{a1}				9.66^{c2}				9.57^{2}

* Reported as mg/dl.

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.002$).

[a] Values with different superscripts within periods are significantly different ($P < 0.04$).

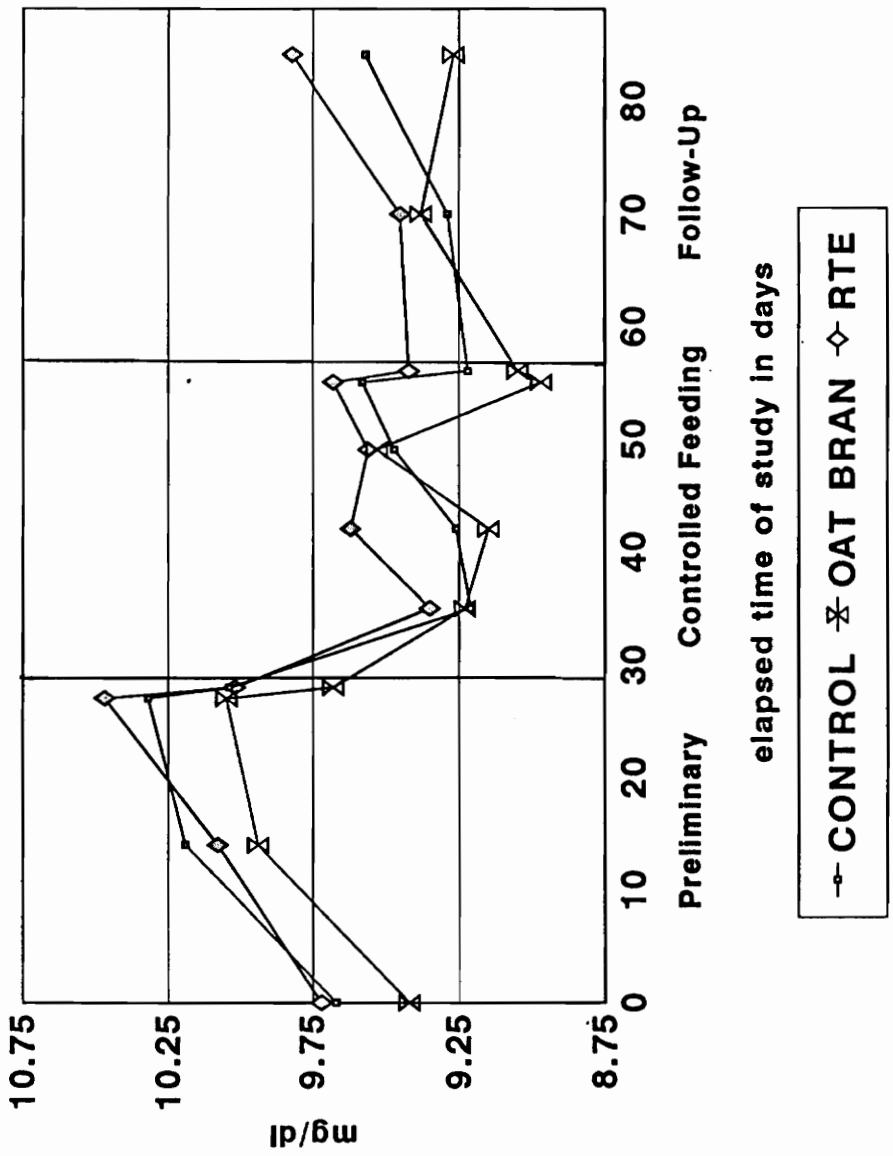


Figure 4 Mean Plasma Calcium

bran group significantly lower than either the control or RTE groups ($P < 0.04$). During the controlled feeding period the RTE group had the highest pooled mean concentration of plasma calcium, the control group intermediate and the oat bran group the lowest ($P < 0.04$). However, by the follow-up period none of the groups showed any significant between group differences in pooled mean plasma calcium concentration. All mean data were within the reported normal plasma concentration range of 9 to 11 mg/dl (Martin et al, 1981).

Magnesium

Mean weekly plasma magnesium data are shown in Table 27 with individual data listed in Appendix Y. Graphic representation of mean plasma magnesium over elapsed time of the study in days is shown in Figure 5. Mean weekly plasma magnesium data ranged from 1.59 ± 1.0 to 1.86 ± 1.3 mg/dl with individual data ranging from 1.44 to 2.06 mg/dl. When fitted to linear, quadratic and cubic models plasma magnesium failed to show any linear, quadratic or cubic effects. After testing for model fit mean plasma magnesium data for each period of the study (preliminary, controlled feeding and follow-up) were pooled and ANOVA statistics performed on the pooled means. Plasma magnesium data for

Table 27

Mean Plasma Magnesium Values of Subjects Consuming Two Types of Oat Bran*

	Period										
	Preliminary				Controlled Feeding					Follow Up	
Group	9/20	10/04	10/17	10/18	10/25	11/01	11/08	11/14	11/15	11/29	12/13
Control	1.72	1.73	1.79	1.82	1.59	1.67	1.75	1.67	1.76	1.69	1.74
± SD	0.3	0.7	1.1	0.9	1.0	0.6	0.9	0.8	0.4	1.0	1.2
			1.77[1]				1.69[a2]				1.71
Oat Bran	1.71	1.72	1.81	1.80	1.66	1.71	1.80	1.72	1.79	1.81	1.75
± SD	1.4	1.0	1.1	1.1	0.9	0.7	0.8	1.5	1.3	0.9	1.2
			1.78				1.76[ab]				1.73
RTE	1.74	1.75	1.84	1.82	1.69	1.71	1.75	1.86	1.80	1.72	1.78
± SD	1.6	1.2	1.2	0.8	1.0	0.9	1.2	1.3	1.3	1.4	1.1
			1.79				1.76[b]				1.75

* Reported as mg/dl.

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.002$).

[a] Values with different superscripts within periods are significantly different ($P < 0.009$).

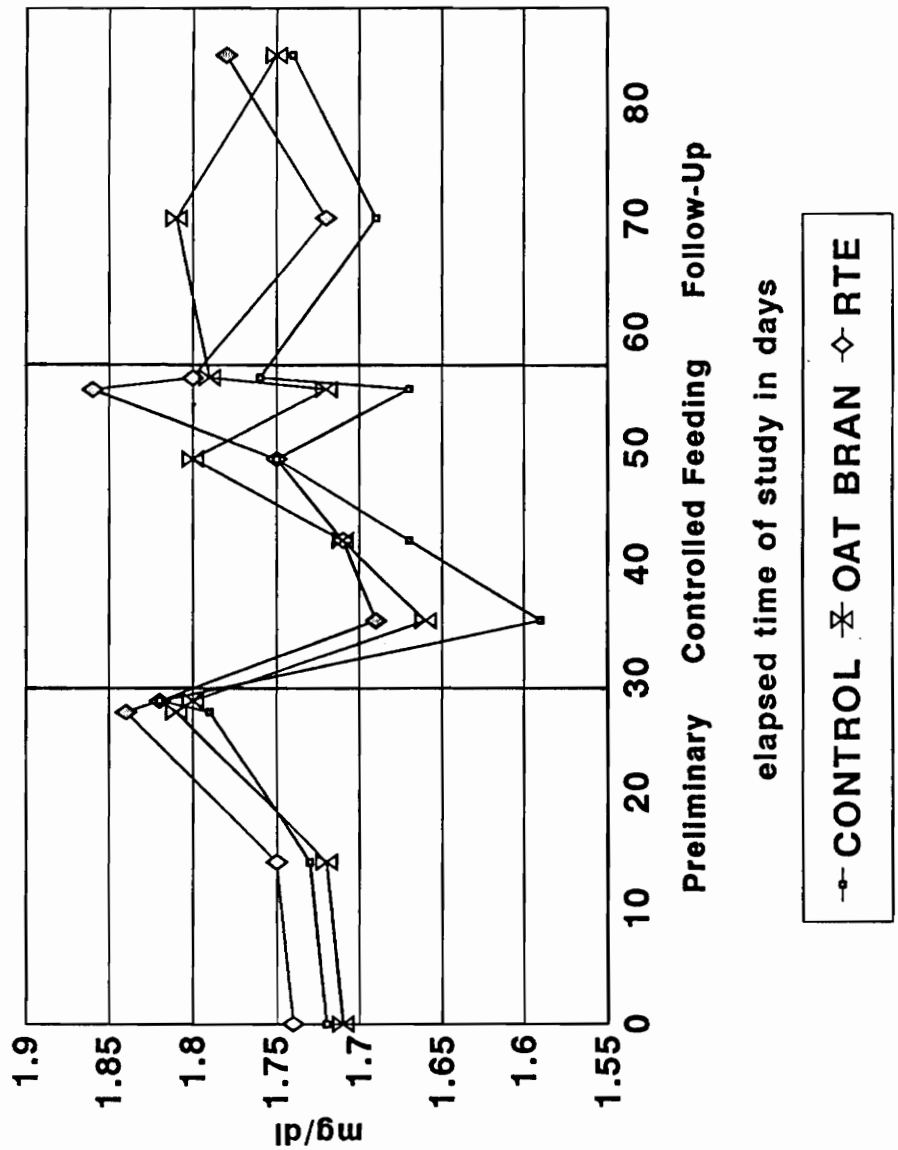


Figure 5 Mean Plasma Magnesium

the control group were significantly higher during the preliminary period than for the controlled feeding period ($P < 0.002$). Both of the fiber supplemented groups failed to show any within group differences during any of the three periods. A between group significant difference was found for the controlled feeding period with the control group's pooled mean plasma magnesium concentration less than the RTE group ($P < 0.009$). All mean data were slightly lower than the reported normal plasma concentration range of 2 to 3 mg/dl (Martin et al, 1981).

Zinc

Mean weekly plasma zinc data are shown in Table 28 with individual data listed in Appendix Z. Graphic representation of mean plasma zinc over elapsed time of the study in days is shown in Figure 6. Mean weekly plasma zinc data ranged from 87.6 ± 9.4 to 123.6 ± 8.2 µg/dl with individual data ranging from 69.3 to 137.0 µg/dl. When fitted to linear, quadratic and cubic models plasma zinc failed to show any linear, quadratic or cubic effects. After testing for model fit mean plasma zinc data for each period of the study (preliminary, controlled feeding and follow-up) were pooled and ANOVA statistics performed on the pooled means. The control and oat bran groups did not show

Table 28

Mean Plasma Zinc Values of Subjects Consuming Two Types of Oat Bran*

	Period										
	Preliminary				Controlled Feeding					Follow Up	
Group	9/20	10/04	10/17	10/18	10/25	11/01	11/08	11/14	11/15	11/29	12/13
Control	102.6	89.7	117.8	108.3	110.4	99.6	103.1	108.9	98.7	95.3	111.4
± SD	7.9	16.2	16.0	10.2	11.3	7.9	9.1	8.3	8.0	9.7	6.4
			104.2[a]				104.1[a]				104.0[a]
Oat Bran	92.1	87.6	110.5	97.2	96.7	99.4	97.9	106.9	97.8	92.5	100.2
± SD	8.1	9.4	9.2	7.1	6.2	8.8	8.7	7.9	4.4	15.5	5.9
			99.2[a]				97.5[b]				96.3[a]
RTE	115.8	102.9	123.6	109.4	104.6	106.3	102.5	114.1	104.8	115.0	112.6
± SD	7.0	13.9	8.2	10.4	3.8	7.9	4.5	6.1	4.4	5.6	9.4
			112.8[b1]				106.3[a2]				113.7[b1]

* Reported as μg/dl.

[1] Values with different numbers within the same dietary treatments between periods are significantly different ($P < 0.03$).

[a] Values with different superscripts within periods are significantly different ($P < 0.03$).

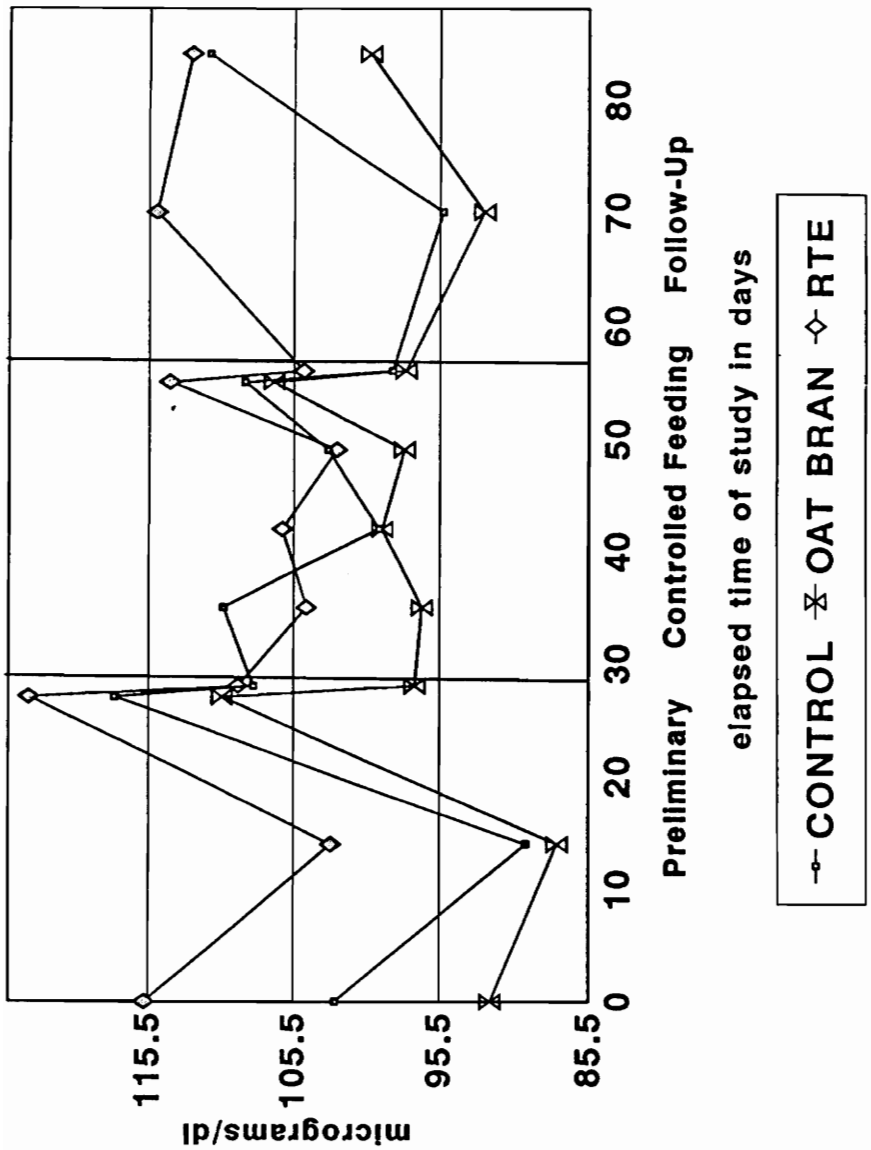


Figure 6 Mean Plasma Zinc

any within group significant differences between the preliminary, controlled feeding or follow-up periods. The RTE group had significantly higher plasma zinc concentrations during the preliminary and follow-up periods than during the controlled feeding ($P < 0.03$). A between group significant difference was found for the preliminary and controlled feeding periods with the oat bran group's pooled mean plasma zinc concentration less than the control and RTE groups ($P < 0.03$). By the follow-up period the control and oat bran groups were both significantly lower than the RTE group ($P < 0.03$). All mean data were within or slightly higher than the reported normal plasma concentration range of 60 to 100 μg/dl (Martin et al, 1981).

Copper

Plasma copper could not be analyzed due to insufficient plasma available.

DISCUSSION

A. Digestibility of Fiber

1. NDF

The control treatment was a basal diet of low fiber foods. The fiber supplemented groups had, in addition to the basal diet, 100 g of either unprocessed or processed (RTE) oat bran cereal. The addition of oat bran fiber, regardless of processing treatment, significantly increased NDF intakes for the fiber supplemented groups. The oat bran group consumed the highest, the RTE intermediate and the control group the lowest amount of NDF. This may have been due to the type of processing treatment that the cereal under went since the unprocessed oat bran group consumed consistently higher amounts of NDF than the processed RTE group. Chang and Morris (1990) reported that heat treatments (processing) in the form of microwaving or autoclaving caused decreases in total dietary fiber and insoluble dietary fiber in oat bran but not of soluble fiber. Shinnick et al. (1988) stated that processing of oat bran to produce high fiber oat flour increased the proportion of fiber recovered as soluble fiber and decreased the insoluble β-glucan fraction. In the current study, the

processing used to produce the experimental RTE oat bran cereal could have affected it's insoluble fiber content. Moak et al. (1987) reported on NDF intakes of subjects consuming high fiber diets based either on wheat bran bread or oat bran bread. During the High Fiber period, their results showed that the oat bran bread group consumed 24.2 g of NDF per day in addition to that supplied by the vegetarian basal diet (38.5 g/day) for a total NDF intake of 62.7 g/day. They failed to report how many slices of the oat bran bread or wheat bran bread their subjects consumed during the High Bran period. Their total dietary intakes of NDF were slightly higher than the NDF intakes seen in this investigation (50.2 g/day oat bran group and 41.1 g/day RTE group in Period I and 43.7 g/day oat bran group and 39.8 g/day RTE group in Period II).

The within group differences seen between the two experimental periods in the current study could possibly be attributed to several reasons. There could have been human errors in compositing or calculating total dietary fiber intakes even though the dietary compositing was strictly supervised and double checked for accuracy by the investigators. The analysis for NDF involves the precise weighing of very dry, small particulate matter, either diet or fecal, and error could have been introduced through the

analysis itself. Differences in moisture content of the dried diets may have contributed to the differences seen. Also, to calculate total dietary intake, the number of supplement units had to be counted daily and included along with the dietary menu intake. Possible miscalculations may have occurred resulting in small errors even though the supplement unit was designed to contain low amounts of dietary fiber. Exactly why Period I values of total NDF intakes were higher than Period II are difficult to explain.

Fecal NDF analysis showed no significant differences among the treatment groups during Period I. During Period II, the oat bran group excreted significantly more NDF than the control. All three groups were excreting only about 8.6 g/day NDF residue per experimental period (average of all three groups). Moak et al. (1987) reported fecal NDF excretions during the High Bran period of 21.4 g/day (consuming 62.7 g/day) for the oat bran bread group, which are larger than those seen in this investigation. During the Low Bran period, Moak et al. (1987) stated the oat bran bread group's fecal NDF excretion of 13.1 g/day which was still above the highest excretion seen in the present study (oat bran group excreted 12.3 g/day during Period II).

The fiber supplemented groups showed a trend of a lower NDF breakdown than the control group, which was consistent

across both experimental periods. Only during Period II did the oat bran group have a significantly lower NDF breakdown than the control group. Slavin et al. (1981) and Marlett and Johnson (1985) both disclosed similar, though non-significant, variable NDF digestibilities in human subjects consuming diets with added cellulose or wheat bran. Their results were in agreement with this investigation in findings that an individual subject's digestibility of fiber is often highly variable. Slavin et al. (1981) suggested that a possible source of within subject variation in fiber digestibility may be due to the subject's ability to adapt to a particular diet or for the colonic microflora to adapt to a particular dietary fiber.

Kivistö et al. (1986) reported that undigested food components were completely excreted in the ileostomy contents on the day of consumption for ileostomy patients consuming a high fiber extruded cereal product. This indicated that if fiber intake was held constant then an adaptation phase was not necessary for fiber consumption. But Kivistö et al. (1986) do not address the issue of colonic microflora's role in fiber digestion since their subjects were all ileostomy patients.

Microflora adaptation to a particular dietary fiber influences fiber digestion. The adaptation time in the

current experiment was eight days prior to the start of the first balance collection. The same subjects remained on their respective diets for the entire duration of the controlled feeding phase resulting in the subjects being fed their diets for twenty days before the second balance collection period began. Adaptation by the microflora in the colon to the diets probably resulted in the significant differences seen between the oat bran group and the control during Period II.

The procedure used in this study, the enzymatic NDF method, removed starch which could cause interference and provides a good estimate of total dietary fiber in cereal products (Lanza and Butrum, 1986; Robertson and Horvath, 1993). For fecal analysis, the enzymatic step for starch removal could be deleted since most all starch present in dietary intakes were either digested or fermented by colonic bacteria. This method has some draw backs, though. The NDF procedure fails to include water-soluble fibers (the fiber material which dissolves in water at 100°C and precipitates in alcohol) (Mongeau and Brassard, 1986). Soluble fibers are lost during extraction with the detergents. This important point is significant when the fiber source used contains large amounts of soluble fiber as is the case with oat products (Lanza and Butrum, 1986; Robertson and Horvath,

1993). Frølich and Nyman (1988) stated that the oat kernel has a dietary fiber content of 11.5 g/100 g and of this, 23% is made up of soluble fiber components, mainly β-glucans. Oat bran is considered a good source of soluble fiber, the major portion of which is β-D glucan, a water-soluble hemicellulose (Seibert, 1987; Klopfenstein, 1988). Beta glucans can be further divided into soluble and insoluble fractions. The oat fiber used in the current study was reported to contain about 25% dietary fiber of which about 1/3 is β-glucan (Quaker Oats Co., 1981). Considering the soluble fiber content of the experimental oat bran used, the NDF procedure may have under-estimated the dietary fiber content. Soluble dietary fiber, which is primarily pectic substances and soluble hemicelluloses, though not recovered by the NDF procedure, are normally totally digested (Marlett and Johnson, 1985; Robertson and Horvath, 1993). These soluble dietary fibers have important physio-chemical functions which have been identified in treating certain disease states such as hypercholesterolemia and diabetes (Anderson and Gustafson, 1988; Whyte et al., 1992; Braaten et al., 1991; Del Toma et al., 1988; Vollendorf and Marlett, 1991).

Another difficulty in NDF determination are problems experienced in filtering the extracted NDF product. High

protein, lipid and starch contents are known to cause filtering problems (Robertson and Horvath, 1993). In an attempt to avoid starch interference a heat stable α-amylase (derived from Bacillus subtilis) was added to breakdown free starch. The α-amylase was determined to be active prior to NDF analysis but could have had it's activity decreased due to the detergents used. Beta-glucans tend to form sticky, viscous solutions when mixed with water which are also difficult to filter (Wood et al., 1989). During this study filtering problems for both NDF and ADF procedures were noted with the formation of gelatinous material around the extracted fiber and could possibly be attributed to the high content of β-glucans present. The β-glucan content of the experimental fibers was not analyzed so the exact cause of the filtering problems is only speculative.

2. ADF

Dietary intakes of ADF were increased due to oat fiber supplementation regardless of processing treatment during Period I. But in Period II, all three groups were consuming similar amounts of ADF. Differences seen between Period I and Period II are not as large as those seen for NDF. In contrast, Moak et al. (1987) reported total ADF intakes for the oat bran bread group during the High Bran period of 35.5

g/day and of 32.4 g/day during the Low Bran period. Their results are considerably higher than those seen in the current investigation but are based on a vegetarian diet.

Fecal ADF for the unprocessed oat bran group was significantly higher than the control treatment during both experimental periods. All fecal ADF excretions were relatively constant over both experimental periods. In comparison, Moak et al. (1987) reported fecal ADF excretions of 10.7 g/day which was higher than that seen for the fiber supplemented groups in the current study (unprocessed oat bran group averaged 6 g/day and processed RTE group averaged 4.3 g/day).

The ADF fraction of fiber contains approximately all of the lignin and cellulose present while removing more than 95% of the hemicellulose present (Robertson, 1976; Goering and Van Soest, 1970; Southgate et al., 1993). Lignin and cellulose are known to be only slightly digested in humans (Van Soest, 1978a; Campbell et al., 1979; Slavin, Brauer and Marlett, 1981). The ADF digestibilities in this study indicate that digestibility was decreased in subjects who consumed diets with added oat bran, especially the unprocessed oat bran. ADF digestibility was significantly decreased for the oat bran group from the control group during both experimental periods. The high digestibilities

seen in this study may result from the type of cellulose contained in the diets which could have been more easily broken down. Since actual cellulose composition was not analyzed, it is unknown why the ADF digestibilities were so high.

In order to estimate the water insoluble hemicellulose content of the diets the ADF fraction was subtracted from the NDF fraction. The estimated hemicellulose content followed the pattern of the dietary NDF content and results showed that the oat bran group consumed the highest amount, the RTE group intermediate and the control consumed the lowest amount of estimated hemicellulose. This pattern was the same during both experimental periods. The oat bran diet contained more NDF than the control or RTE diet, therefore, the oat bran diet contained more estimated hemicellulose.

B. Minerals

Analyzed intakes of calcium and magnesium were above the recommended allowances of the Food and Nutrition Board of the National Academy of Sciences (1989), while zinc was below the RDA, and analyzed intakes of copper were within the estimated safe and adequate range (ESADDI). The diets fed in this study were not supplemented in any way other

than the addition of the unprocessed or processed oat bran fiber so that any mineral alterations could be attributed to the experimental fibers in question. Oats are reported to contain ample amounts of calcium, magnesium, zinc, and copper (Peterson, 1992). The inclusion of oat bran in this study appeared to increase the dietary intakes of magnesium, zinc, and copper regardless of processing treatment. These findings are in agreement with those of Moak et al. (1987) and Spencer et al. (1991) that increased consumption of oat fiber resulted in increased magnesium, zinc, and copper intakes. In the current study, the oat bran group appeared to have a decreased intake of calcium during both experimental periods from either of the other two treatment groups. This is most likely due to the menu composition and the fact that on each day of the controlled feeding period the control group consumed 141 g of 1% fat cottage cheese which the other two groups did not consume (Table 11). But, the oat bran and RTE groups still consumed over the RDA for calcium. The subjects in both Moak et al. (1987) and Spencer et al. (1991) were supplemented with calcium as calcium carbonate or calcium gluconate tablets, respectively. The calcium intakes reported by Moak et al. (1987) with supplementation were still below the RDA level while calcium intakes reported by Spencer et al. (1991) were

only slightly above the RDA.

Oat bran supplementation in the present study had no effects on urinary excretion of calcium, magnesium, or zinc. Krause and Mahan (1976) state that urinary losses of calcium, magnesium, and zinc are relatively constant over a wide range of mineral intakes. These results were consistent with results from most studies involving dietary fiber. Studies with wheat bran showed no significant effects on urinary excretion of these minerals (Sandstead et al., 1978; Sandberg et al., 1982; Andersson et al., 1983). Urinary magnesium has been shown to increase when human subjects were fed wheat bran (Reinhold et al., 1976; Rendleman et al., 1982; Van Dokkum et al., 1982). In studies looking at hemicellulose as a fiber source similar results showed that increased hemicellulose content had no effect on urinary calcium, magnesium, or zinc excretion (Kies, Fox and Beshgetoor, 1979; Drews, Kies and Fox, 1979).

In animal studies also involving oat fiber, the indications were also that oat fiber had no apparent effect on urinary calcium, magnesium, or zinc excretion (van der Aar et al., 1983; Shah et al., 1990). Human studies involving oat fiber have indicated similar findings to those results found in the current study. The results from the current study are in agreement with published findings that

urinary zinc was not affected by oat bran supplementation but are inconsistent with published reports concerning urinary calcium and magnesium excretions (Moak et al., 1987; Spencer et al., 1991). Moak et al. (1987) found that urinary zinc and calcium were unaffected by oat bran supplementation but that urinary magnesium was significantly increased. Spencer et al. (1991) found in humans that urinary calcium excretion decreased significantly, that urinary magnesium increased and that urinary zinc excretion was unchanged.

Oat fiber supplementation had significant effects on fecal mineral excretions. Fecal magnesium and zinc excretions were significantly increased for both fiber supplemented groups over the control, regardless of processing treatment. Fecal calcium and copper excretions were significantly increased for the processed RTE oat bran group over either the unprocessed oat bran or control groups. These results agree with published findings involving added wheat bran that fecal calcium, magnesium and zinc excretions were significantly increased (Reinhold et al., 1976; Sandstead et al., 1978; Van Dokkum et al., 1982).

Spencer et al. (1991) found that the consumption of oat bran muffins caused non-significant changes in fecal calcium excretion but fecal magnesium and zinc excretions were

significantly increased. Moak et al. (1987) reported that oat bran supplementation in the form of bread resulted in significantly increased fecal excretions of calcium, magnesium, and zinc. Fecal copper excretions were increased, but only due to increased consumption of copper and not apparently due to the type of fiber consumed. Balasubramanian et al. (1987) found significantly increased fecal calcium losses in older subjects supplemented with 30 g of wheat bran.

It appears that as the intake of minerals increased, fecal excretion can also increase. Magnesium absorption, for example, varied inversely with intake: when intake was low absorption was high and vice versa (NRC, 1989). Often fecal mineral losses correlate with daily dietary intakes (Krause and Mahan, 1976). If this was the case then the RTE group which had the highest intake of calcium would in turn have a high level of fecal calcium excretion. The experimental fiber may have had an effect on fecal calcium excretion but the tests employed in this study were not sensitive enough to determine what that effect may have been. Likewise, high intakes of magnesium and zinc would in turn be associated with high fecal excretions of these minerals. However, this idea does not hold true for copper since both fiber supplemented groups consumed near equal

amounts of copper but only the RTE group excreted significantly more than the control. The processing treatment of the oat fiber may have had some effect on fecal mineral losses, particularly for calcium and copper since a significant difference was seen for one fiber group over the other. The processed RTE group excreted more calcium and copper than either the control or unprocessed oat bran groups. When interpreting calcium results it must be remembered that the RTE group was consuming the highest amount of calcium and the oat bran group the lowest. If increased intakes were reflected in increased fecal output then it would be logical that the RTE group should have been excreting more calcium than the oat bran group.

This assumption of increased intake resulting in increased fecal excretion is difficult to apply to fecal copper excretion since both fiber supplemented groups were consuming nearly equal levels. Therefore, a possible processing effect may have been contributing to this difference in copper excretions. During processing, the RTE cereal was heated in order to produce the desired textured product. Heating of cereal products tends to decrease the phytate level (Caprez and Fairweather-Tait, 1982; Frølich, 1993), while increasing the formation of artifact lignin via the Maillard reaction (Robertson, 1976). Lignin is known to

bind minerals reducing availability. In an in-vitro study of the binding characteristics of lignin, cellulose, guar gum, pectin and NDF fiber, Platt and Clydesdale (1987) found that lignin had two high affinity binding sites for iron, copper, and zinc in that order, but with twice as much copper bound as either iron or zinc. Toasting of wheat bran has been found to significantly affect mineral binding most likely due to the small amount of lignin formed (Camire and Clydesdale, 1981). Processing to manufacture the RTE oat cereal used in this study may have slightly increased the lignin content resulting in increased binding of copper. Processing can also reorganize dietary fiber components changing the chelating properties of the components (Frøhlich, 1993). Kivistö et al (1986) found that when subjects were fed a processed (extruded) wheat bran fecal excretions of calcium, magnesium, and zinc were increased above excretions seen for the same subjects receiving a non-processed wheat bran. This agrees with the results of the current investigation that the processing treatment of oat bran can affect fecal excretion of calcium, magnesium, zinc, and copper. It is difficult to determine if the effects seen are primarily due to the processing treatment, other factors, or due to the fact that oat bran increases dietary intakes of minerals resulting in increased excretions.

Another possible mechanism for increased fecal losses of minerals could be due to the nature of soluble dietary fibers, especially β-glucans, to form gels or viscous solutions when mixed with water (Wood, 1986; Klopfenstein, 1988; Wood et al., 1989). This formation of gels may trap available minerals and displace them away from the lumenal wall resulting in decreased absorption and increased excretion. Vollendorf and Marlett (1991) disclosed that soluble fiber was the major factor responsible for oat fiber's ability to lower serum cholesterol levels, possibly by this same gel forming feature.

Results indicate that oat bran supplementation, regardless of processing treatment, had no effects on apparent calcium, zinc, and copper retentions. The unprocessed oat bran fiber appeared to have a significant effect on apparent positive magnesium retention during Period I but this significance was lost by Period II. Apparent calcium balances were negative for the RTE group during both experimental periods and for the oat bran group during Period II. The mean apparent mineral balances for magnesium, zinc, and copper were positive for all groups during both periods with the exception of zinc for the control group during Period I. Individual subject's apparent mineral balances were highly variable resulting in

large standard error of the means.

Balances or retentions were reported as apparent balances since no attempt was made to collect sweat or dermal losses or to estimate possible endogenous sources. These results are in agreement with most studies involving supplemented dietary fibers. In studies using wheat bran, apparent mineral balances of calcium and magnesium were unchanged due to the addition of wheat bran (Reinhold et al., 1976; Sandberg et al., 1982; Van Dokkum et al., 1982); while studies looking at zinc and copper indicated that zinc balance can be negatively affected and copper balances positively affected by the addition of wheat bran (Sandstead et al., 1978; Sandberg et al., 1982; Kies, Fox and Beshgetoor, 1979). Several studies stated that apparent balances for calcium, magnesium, zinc, or copper were negative when subjects consume high intakes of wheat bran or fiber from fruit and vegetable sources (Iamail-Beigi et al., 1977; Kelsay et al., 1988). Spencer et al. (1991) reported negative apparent balance of calcium and positive balances of magnesium and zinc. Moak et al. (1987) reported negative apparent balances of copper, zinc, and magnesium and positive apparent balance of calcium during both periods. It would appear that the current findings concerning apparent mineral balance agree with those of Spencer et al.

(1991), but are inconsistent with those findings of Moak et al. (1987).

The lack of effect of oat bran on mineral metabolism in men in the present study may be due to the high content of soluble fiber in oat bran or to the phytate content. Soluble dietary fibers, especially β-glucans, tend to form sticky viscous gels when mixed with water (Wood, 1988; Klopfenstein, 1988; Wood et al., 1989). These viscous solutions may bind minerals in the small intestine thus rendering the minerals unavailable for absorption. However, it must be remembered that soluble fibers are fermentable by colonic microflora (Marlett and Johnson, 1985; Robertson and Horvath, 1993) which may result in the release of the bound minerals in the colon. It is unclear if these released minerals would be absorbed in the colon by the human or used by the microflora themselves. Calcium is known to be absorbed into the body in a number of ways including passive diffusion if a concentration gradient is present (Krause and Mahan, 1976; Martin et al., 1981).

Processing of the oat bran may have some role in the lack of affect on apparent mineral retention. The unprocessed oat bran used was presumed to have been processed only in so far as to deactivate lipases to prevent rancidity. This short term "gentle heating" may not have

been long enough to deactivate the phytases present. Phytates are known to complex divalent minerals to form insoluble salts which are unabsorbable (Davies, 1979; Krause and Mahan, 1976). But published results are inconclusive as to effects, if any, that phytate may have on mineral availability in the human. Turnlund et al. (1984) found that phytate greatly reduced zinc absorption while Morris and Ellis (1983); Ellis et al. (1987); Forbes et al. (1983); Forbes et al. (1984); and Mason et al. (1990) found no effect of phytate on zinc availability in humans. The dietary fiber of oats is a mixture of soluble and insoluble fractions with a high soluble fraction relative to other cereals (Peterson, 1993). This soluble fiber fraction appears to be mainly β-D-glucans and water soluble hemicelluloses (Klopfenstein, 1988). Phytate is also found in this soluble fiber fraction (Frølich and Nyman, 1988). Rendleman (1982) reported from in-vitro studies that water-soluble components are responsible for more than half of the binding ability of wheat bran, and the principle soluble chelating agent is phytate.

The experimental fibers used in this investigation were of considerable β-glucan content (one third) and this could possibly cause mineral binding. Studies by Wood et al. (1989) indicate that the soluble fiber fraction of oats was

physiologically active. Oat β-glucans are enriched in the bran fraction of oats and are therefore not lost during milling. The unprocessed oat bran used in this study was obtained by passing milled oat through a screen to produced a bran rich fraction retained on the screen (Quaker Oats Co., 1981). Filtering problems associated with the NDF and ADF procedures support the idea that the experimental oat bran contained a considerable amount of β-glucan. Neither β-glucan, nor phytate, nor soluble fiber were analyzed for in this investigation making interpretation only speculative at this point.

Researchers have begun questioning the length of adaptation needed for interpreting mineral changes due to dietary influences. Studies by Schwartz et al. (1986) and Kelsay et al. (1988) concluded that when investigating mineral balances measurements made on a single week cannot be taken as representative of a significant metabolic response no matter how well adapted a subject might be.

> "Significant weekly differences in mineral balances may be important indicators in the validity of interpretation of balance data. Response to a dietary regimen should be measured for several weeks. Because of variations in balances from week to week it is difficult to interpret balance results, and any given

> week is not sufficient. It is probably best to use the results of several consecutive weeks of balance when interpreting the effects of a certain dietary regimen". (Kelsay et al., 1988)

Schwartz et al. (1986) concluded that at least 4 weeks were needed for adaptation in investigations involving one or more minerals when the experimental diet was adequate in the nutrients under investigation and that measurements of responses to treatment required 2-3 week each. The current metabolic study involved a controlled feeding period of 28 days which was divided into an 8-day adaptation, an 8-day balance period, a 4-day break, with a second 8-day balance period. This time frame is similar to that used by Moak et al. (1987) but not to that used by Spencer et al. (1991). Moak et al. (1987) utilized a 7-day adjustment period followed by a high fiber period of 14 days (balance collections were made during the last 7 days) followed by a low fiber period of 14 days (balance collections were made during the last 7 days). The study by Spencer et al. (1991) involved a 4-week adaption phase, a 40-day control period, and a 32-day experimental period. It is interesting that this study's results agree more with Spencer et al. (1991), a long term metabolic study than with Moak et al. (1987), a short term metabolic study. A study's duration and the

adaptation of subjects to an experimental diet may influence results.

It must be remembered that individual intakes of caffeine containing beverages (coffee and tea) were permitted throughout the course of this study. Contradictory results have been published concerning coffee, tea, or caffeine's possible affects on mineral excretions and balances in humans (Massey and Hollingberry, 1988; Barger-Lux et al., 1990). The subject's consumption of these beverages needs to be considered when interpreting the results of this investigation but their effect would be unlikely since no unusually high consumptions were noted and no significant effects on balance were seen for any of the minerals in question except for Period I magnesium. Massey and Hollingberry (1988) found that only urinary calcium was affected by caffeine consumption, while Barger-Lux et al. (1990) found no effect on urinary calcium excretion. The current results indicated that there was no effect of coffee, tea, or caffeine on urinary calcium. And coffee, tea or caffeine probably had no apparent affect on mineral balances in the adult male subjects. Future studies concerning mineral balances need to have these dietary limitations eliminated, if possible, to avoid confounded results.

C. Plasma Mineral Concentrations

In interpreting the plasma mineral concentrations the data was fitted to linear, quadratic and cubic models. None of the analyzed minerals showed any linear, quadratic or cubic effects. To test for within group or between period significant differences the analysis of variance with contrasts model was employed.

Various researchers have reported that increased consumption of various dietary fibers had no affect on certain plasma mineral concentrations (Mason et al., 1990; Heaton et al., 1976; Ismail-Beigi et al., 1977; Kies et al., 1979; Drews et al., 1979; Spencer et al., 1991). A few studies indicated possible decreases in plasma calcium concentrations in subjects fed wheat bran (Heaton and Pomare, 1974; Ismail-Beigi et al., 1977). Plasma calcium, magnesium, and zinc concentrations appear not to be correlated with dietary intake (Kant et al., 1989).

Within treatments across the study periods (preliminary, controlled feeding, and follow-up) plasma calcium concentrations were significantly lower for all groups during the controlled feeding and follow-up periods than during the preliminary period.

Plasma magnesium concentrations were higher for the control group during the preliminary period than for the

controlled feeding period. The fiber supplemented groups failed to show any differences in plasma magnesium during any of the periods.

Plasma zinc levels for the RTE group were significantly higher during the preliminary and follow-up periods than for the controlled feeding phase. The control and oat bran groups did not show any significant differences in plasma zinc concentrations during any of the periods.

Comparing across groups during the study periods, results showed that during all three periods the RTE group had the highest mean plasma levels of calcium and zinc, the control group intermediate and the oat bran group the lowest mean plasma values. Mean plasma magnesium concentrations were consistent across all three divisional periods with only a decrease seen for the control group during the controlled feeding.

All plasma mineral concentrations tested were within or only slightly lower than reported normal plasma mineral ranges (Martin et al., 1981). Plasma mineral results from this study agree with findings of Spencer et al. (1991) that oat fiber supplementation had no effect on plasma calcium, magnesium, or zinc levels.

SUMMARY AND CONCLUSIONS

A metabolic diet study was conducted to determine the effect of unprocessed and processed oat bran fiber on calcium, magnesium, zinc, and copper intakes, excretions, apparent balances, and plasma mineral concentrations in adult men. Twenty-one subjects were recruited from the faculty and graduate student populations at Virginia Tech to participate in the feeding study. All subjects were deemed in good health by their physician and were given both oral and written explanations of the study. The study followed the guidelines of and was approved by the Institutional Review Board of Research with Human Subjects at Virginia Tech.

The subjects were randomly assigned to one of three treatment groups: (1) basal diet with no added fiber - control; (2) basal diet with 100 g of unprocessed oat bran - oat bran; or (3) basal diet with 100 g of processed ready-to-eat (RTE) oat bran cereal - RTE. The study consisted of a preliminary, a controlled feeding, and a follow-up period of four weeks each. During the preliminary period the subjects completed dietary recalls and questionnaires concerning exercise levels and medication usage. The controlled feeding period was subdivided into an 8-day

adjustment period followed by an 8-day balance period (Period I), a 4-day break in collection, followed by a second 8-day balance period (Period II). Throughout the balance periods the subjects collected all urine and fecal excretions in containers supplied by the investigators. A four day menu cycle was used and food samples were collected and composited into 4-day composites. Urine was composited daily and pooled for each 8-day balance period. Feces were collected and frozen until the end of the study at which time they were pooled and composited for each 8-day balance period. Fasting blood samples were drawn biweekly during the preliminary and follow-up periods and weekly during the controlled feeding phase. Diets were analyzed for nitrogen, fat, and moisture following A.O.A.C. (1975) procedures; for neutral detergent (NDF) and acid detergent (ADF) fiber by the procedures of Robertson and Van Soest (1977); and for mineral concentration by wet oxidation with absorbencies determined by atomic absorption spectrophotometry (Colin et al., 1983). Fecal fiber, urinary and fecal minerals, and plasma minerals were analyzed similar to the diets.

The addition of oat bran, regardless of processing treatment, increased NDF intakes for the fiber supplemented groups. The unprocessed oat bran group consumed the highest, the processed RTE group intermediate, and the

control group the lowest amount of NDF. Differences may possibly be attributed to the type of processing treatment that the fiber under went since the unprocessed oat bran group consumed consistently higher amounts of NDF than the processed RTE group. Fecal NDF analysis showed no differences among the treatment groups during Period I, but during Period II, the oat bran group excreted more NDF than the control group. The fiber supplemented groups showed a trend toward decreased NDF breakdown which was consistent across both experimental periods. Only during period II was this decrease significant with the oat bran group lower than the control.

Dietary intakes of ADF were increased due to oat fiber supplementation regardless of processing treatment during Period I; but in Period II all three groups were consuming similar amounts of ADF. Fecal ADF for the unprocessed group was significantly higher than the control treatment during both experimental periods. All fecal ADF excretions were relatively constant over both experimental periods. ADF digestibilities in this study indicated that digestibility was decreased in subjects who consumed diets with added unprocessed oat bran but only during Period II.

In order to estimate the water insoluble hemicellulose content of the diets the ADF fraction was subtracted from

the NDF fraction. The estimated hemicellulose content followed the pattern of the dietary NDF content and results showed that the oat bran group consumed the highest, the RTE group intermediate and the control consumed the lowest amount of estimated hemicellulose. This pattern was the same during both experimental periods.

The inclusion of oat bran in this study appeared to increase the dietary intakes of magnesium, zinc, and copper regardless of processing treatment. Analyzed intakes of calcium and magnesium were above the recommended allowances of the Food and Nutrition Board of the National Academy of Sciences (1989), while zinc was below the RDA, and analyzed intakes of copper were within the estimated safe and adequate range (ESADDI). The diets fed in this study were not supplemented in any way other than the addition of the unprocessed or processed oat bran fiber so that any mineral alterations could be attributed to the experimental cereals in question. The oat bran group appeared to have a decreased intake of calcium during both experimental periods from either of the other two treatment groups. This was most likely due to the menu composition and the fact that on each day of the controlled feeding period the control group consumed 141 g of 1% fat cottage cheese which the other two groups did not consume. However, the oat bran and RTE

groups still consumed more than the RDA for calcium.

Oat bran supplementation had no effect on urinary excretion of calcium, magnesium, or zinc for any of the experimental groups. Oat fiber supplementation had significant effects on fecal mineral excretions. Fecal magnesium and zinc excretions were significantly increased for both fiber supplemented groups over the control, regardless of processing treatment. Fecal calcium and copper excretions were significantly increased for the processed RTE oat bran group over either the unprocessed oat bran or control groups. Increased intake of the minerals appeared to contribute to the increased fecal losses, although, the processing treatment of the oat bran may have affected fecal copper excretion.

The data of the present study suggested that oat bran supplementation had no effect on apparent calcium, zinc, and copper retentions, regardless of processing treatment. The unprocessed oat bran fiber appeared to have a significant positive effect on apparent magnesium retention during Period I, but this significance was lost by Period II.

All plasma mineral concentrations were within or only slightly lower than reported normal plasma mineral ranges. Plasma mineral concentrations appeared to be significantly lower during the controlled feeding period than during the

preliminary and follow-up periods for calcium and zinc. The addition of unprocessed oat bran resulted in a significant decrease in plasma calcium and zinc while plasma magnesium levels were unaffected.

From this study involving oat bran fiber it can be concluded that the addition of 100 g of oat bran, regardless of processing treatment, did not significantly affect calcium, magnesium, zinc or copper apparent retentions in adult men. Oat bran appeared to be a good dietary source of magnesium, zinc, and copper. Increased intakes of these minerals appeared to contribute to the increased fecal losses. The addition of oat bran caused significantly increased intakes of neutral detergent fiber and estimated hemicelluloses. Plasma mineral levels were affected by the addition of oat bran, but levels stayed slightly below or within the normal ranges.

Much information on mineral bioavailability from unprocessed and processed oat bran fiber consumption by adult men was derived from this study. In order to substantiate the results herein further studies on the oat bran used are needed. Dietary fiber is such a complex entity that it is difficult to relate any mineral bioavailability results directly to the fiber in question. Fiber fractionation into insoluble and soluble components

may provide additional insight into oat bran's total dietary fiber content. Perhaps future work should focus on which fraction of oat fiber is involved in oat bran's gel forming ability and this fraction's mineral binding capacity. In-vitro studies similar to those of Ismail-Beigi, Faraji, and Reinhold (1977) using the two types of oat bran fibers used in this investigation may lend insight into exactly which component(s) of the oat fiber is (are) causing mineral binding. In-vitro studies are less costly than metabolic balance studies and more easily controlled in the laboratory setting.

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APPENDICES

APPENDIX A

Personal Physician Clinical Evaluation Form

I,_____authorize _____, M.D. to release requested information about my health to Forrest W. Thye, Ph.D., Human Nutrition and Foods, VPI & SU.

Signed_____
Signature of Subject

I have recently given a physical examination to and reviewed the health record of _____ and find a/no (circle one) medical condition or physical impairment that precludes his/her participation in the human feeding and metabolic experiment in the Department of Human Nutrition and Foods.

_____ _____, M.D.
Date Signature

APPENDIX B

University Health Services Clinical Record Evaluation Form

I, _____ authorize C.W. Schiffert, M.D., Director of the Virginia Tech Health Service, to release requested information about my health to _____.

Signed _____
Signature of Student

I have reviewed the Virginia Tech Clinical record of_____, and find a/no (circle one) medical condition or physical impairment that precludes his/her participation in the following activity: _____

_____.

_____ Signed_____
Date Director of Student Health Service

APPENDIX C

Written Statement of Purpose and Design of Experiment

The study of the relation of dietary fiber to coronary heart disease (CHD) is a fairly recent phenomenon. Epidemiological studies first indicated that populations consuming high levels of dietary fiber had a low incidence of heart disease. Studies in the U.S. and other western countries using a variety of fiber sources have indeed shown that certain dietary fibers will lower serum and LDL cholesterol. Results vary, however, and studies with oat bran are limited. This research will investigate the effect of two different forms of oat bran (primary source of dietary fiber) on plasma levels of total cholesterol and lipoprotein (HDL, LDL, and VLDL) cholesterol levels, and retention of zinc and copper (possibly other minerals) in men.

Subjects will be divided into 3 dietary treatment groups: control, processed oat bran (RTE) and unprocessed oat bran. Assignment to treatment groups will be by randomized block design, using plasma cholesterol levels to rank the experimental subjects. All three dietary groups will consume approximately 2700 calories. Weight will be kept constant; if you begin to gain or lose weight, your calories will be adjusted.

All diets have been planned by careful calculation of calories, protein, fiber, saturated and polyunsaturated fat and cholesterol. Everything you eat will be composited and analyzed so we know exactly what nutrients you take in. Each week blood samples will be taken to analyze for plasma cholesterol (total, LDL, HDL, and VLDL) levels. Food, fecal and urine composites will be analyzed for zinc, copper, and other minerals to determine their retention.

Now that you have an understanding of the purpose of the study, we want to emphasize the importance of adhering to the dietary regimen prescribed for you throughout the controlled feeding period. Please let us know of any suggestions that you may have regarding the diets, or how we can make things better for you.

Thank you for your interest.

APPENDIX D

Notice of Participation in Oat Bran Study

MEMO

TO: Potential subjects for the dietary fiber feeding project

FROM: Forrest W. Thye, HNF

RE: Volunteering as subjects for the Dietary Fiber Study this fall

DATE: August 4, 1983

We finally received word that we would be funded by Quaker Oats for the feeding project this fall to study the effect of oat bran fiber on serum cholesterol (total and lipoprotein fractions) and steriod excretion. We are presently working on menus for the feeding portion of the project and getting organized in the metabolic unit and laboratories.

Your plasma total cholesterol level was high enough to qualify you for the study and if it has not dropped precipitously since our screening sample, we would very much like to consider you for the project.

Of course we need to know as soon as possible if you can help us by being a subject on the project. The following tentative schedule will give you an idea of what would be expected:

September 20	-Blood Sample
September 26-30	-5-Day Dietary Record
October 4	-Blood Sample
October 10-14	-5-Day Dietary Record
October 17 and 18	-Blood Sample
October 18	-Begin Controlled Feeding
October 24-Nov 1	-8-Day Fecal and Urine Collections
October 25	-Blood Sample
November 1	-Blood Sample
November 7-15	-8-Day Fecal and Urine Collections
November 8	-Blood Sample
November 14 and 15	-Blood Samples
November 15	-End Controlled Feeding
November 21-25	-5-Day Dietary Record
November 29	-Blood Sample
December 5-9	-5-Day Dietary Record
December 13	-Blood Sample, End of Study

The most important period and when we must have you in Blacksburg will be during the controlled feeding portion of the experiment, October 17 to November 15. Please make sure that your schedule will allow this. This involves eating three meals a day, 7 days a week for 4 weeks in our facility while eating and drinking only what we provide.

There is a small stipend for each subject completing the project plus free food for 4 weeks. Hopefully this helps in a small way to make up for the inconvenience of being a subject. Obviously, your plasma cholesterol values will be made available to you as well.

Please fill out the form below and return as soon as possible so that we can make plans accordingly.

____ Yes, I can participate. Let me know when the orientation meeting will be.

____ Sorry, I am unable to participate in your study.

____ Sorry, I am unable to participate in your study at this time, but would be interested at another time.

APPENDIX E

Consent of Participation
Nutrition Study

Department of Human Nutrition and Foods

I have received an explanation of the study and understand the following:

As a subject, I will be on a diet during the period from _____ to _____, 1983. I understand that I am to consume only the food and drink provided in the metabolic kitchen. Diets will provide nutrients at levels recommended by the National Research Council, Food and Nutrition Board (1980). The diets will be such that cholesterol levels of 500 mg/day will be consumed.

Venous samples of blood, approximately 40 ml, will be taken at biweekly intervals during the preliminary and follow-up periods and at weekly intervals through the controlled feeding period of the study. A total of no more than 440 ml of blood will be taken over a twelve week period (11 blood samples). Blood samples will be taken by qualified medical personnel. Individual serum cholesterol data will be available to each participant.

Zinc and copper retentions will be determined for two 8-day experimental periods during controlled feeding based on intake and urine and fecal data.

Subjects will be paid $150.00 for completing the feeding and metabolic study and for the preliminary and follow-up blood samples. A subject may drop out of the study for health or personal reasons.

A health physical check will be made prior to the feeding study through the University clinic and personal physicians. If at any time a participant so desires or the investigators or physician believe that the health of the subject may be impaired, the person may drop from the study.

Participants are invited to ask any questions about procedures at any time.

No compensation or medical treatment, other than those normally available through student health services and emergency service by the rescue squad, is available if injury is suffered as a result of this research.I understand the above and agree to participation in the study from _____ through _____ with follow-up blood samples on _____ and _____, 1983.

_____	_____
Telephone	Signature
_____	_____
Date	Local Address

F. W. Thye, Investigator
961-6620
961-5549

C. D. Waring, Chairman
IRB
961-5284

APPENDIX F

OAT BRAN STUDY
FALL 1983
INSTUCTIONS FOR SUBJECTS

MEAL TIMES AND FOOD AND BEVERAGE INTAKE

Meal times: All meals will be served at Solitude fro the entire four week metabolic feeding period of the study. The meal times will be as follows:

	Monday-Friday	Saturday-Sunday
Breakfast:	7:15 - 8:30 (8:45 on blood days)	8:15 - 9:30
Lunch:	11:30 - 1:00	12:00 - 1:30
Dinner:	5:00 - 6:00	5:00 - 6:00

Meal times means that you need to be at Solitude to receive your meal tray within the scheduled times. You do not have to be finished eating by the end of the meal times.

Mealtime Procedures: For the study to run smoothly, it's important to follow the procedures below:

1. Try to arrive at Solitude for each meal at the time you previously indicated.

2. Enter Solitude by the door facing the Duck Pond.

3. Drop off your fecal collection cup in the designated trash can on the porch. (Only during total collection periods.)

4. Place your urine collection bottle in the designated area. (Only during total collection periods.)

5. Tell graduate student supervisor on duty that you have arrived, and tell her what beverage you want with your meal.

6. Have your weight taken and recorded by the graduate

student supervisor. (Before breakfast only)

7. Report to the graduate student supervisor any medication that you took in the past 24 hours. NOTE: Please avoid taking non-prescription medications whenever possible, including aspirin.

8. Wash your hands thoroughly in the second floor bathroom. Do not apply hand cream.

9. Go to the dining area, tell a member of the kitchen preparation staff your name, and pick up your tray from the kitchen.

10. Check the items marked on your tray card against those on your tray. Report any discrepancies to the graduate student supervisor.

11. While eating, use your rubber spatula to clean each dish as completely as possible. Rinse all beverage cups with a small amount of deionized water.

12. When you have finish eating, have a graduate student supervisor check your tray and remove your tray card.

13. Carry your tray from the dining area. Place spatulas and Corning Ware bowls (when used) in the designated pan on the table near the door where you entered.

14. Place all disposable items in the designated trash can on the porch.

15. Place your tray on the table with the spatulas and bowls.

16. Have your water bottle refilled.

17. Pick up clean fecal and urine collection containers. (Only during total collection periods.)

Food and Beverage Consumption Rules:

1. Only food and beverages provided by the study can be consumed. No water, other than the deionized water provided by the study can be consumed.

2. All food must be eaten at Solitude at the designated meal times, unless special arrangements have been made in advance. (This includes occasional meals that must be eaten elsewhere, and designated snacks for those subjects requiring very high caloric intake.)

APPENDIX G

SCHEDULE FOR OAT BRAN DIETARY STUDY

September 20	Blood Sample
September 26-30	5-Day Dietary Record
October 4	Blood Sample
October 10-14	5-Day Dietary Record
October 17-18	Blood Samples
October 18	Begin Controlled Feeding
October 24-Nov 1	8-Day Fecal and Urine Collection
October 25	Blood Sample
November 1	Blood Sample
November 7-15	8-Day Fecal and Urine Collection
November 8	Blood Sample
November 14-15	Blood Samples
November 15	End Controlled Feeding
November 21-25	5-Day Dietary Record
November 29	Blood Sample
December 5-9	5-Day Dietary Record
December 13	Blood Sample, end of study

APPENDIX H

Composition of Experimental Diets[1]

Nutrient	Control Diet	Unprocessed Oat Bran	Processed Oat Bran
		Mean ± SEM	
Energy (kcal)	2862.25±79.03	2677.25±79.03	2677.50±79.03
Protein (g)	86.83± 2.55	79.88± 2.55	79.68± 2.55
Fat (g)	125.20± 7.63	113.28± 7.63	113.48± 7.63
Carbohydrate (g)	356.35±20.04	344.40±20.04	344.32±20.04
Cholesterol (mg)	531.00±41.65	488.50±41.65	488.50±41.65
SFA* (g)	51.43± 4.32	44.03± 4.32	43.93± 4.32
PUFA** (g)	23.90± 1.25	22.93± 1.25	22.93± 1.25
P/S Ratio***	0.48± 0.04	0.52± 0.04	0.52± 0.04
Calcium (g)	1.25± 0.11	0.94± 0.11	0.94± 0.11
Dietary Fiber (g)	4.38± 0.34	19.43± 0.34	20.13± 0.34

[1] Based on computerized analysis of diet menus.

* SFA - Saturated Fatty Acid

** PUFA - Polyunsaturated Fatty Acid

*** P/S Ratio - Polyunsaturated/Saturated Fatty Acid Ratio

APPENDIX I

Mean Percent Recommended Dietary Allowances of Experimental Diets*

Nutrient	Control Diet	Unprocessed Oat Bran Diet	Processed Oat Bran Diet
Protein	155	143	142
Vitamin D	156	139	150
Vitamin E	159	130	150
Vitamin C	457	365	365
Folacin	129	101	101
Niacin	64	51	51
Riboflavin	132	93	96
Thiamin	110	80	80
Vitamin B_6	73	62	62
Vitamin B_{12}	136	97	97
Calcium	157	118	118
Iodine	148	135	135
Iron	116	97	97
Magnesium	93	79	79
Phosphorus	191	142	142
Zinc	57	45	45

* Based on computerized analysis of diet menus.

APPENDIX J

SUBJECT LIST AND INITIAL WEIGHTS FOR OAT BRAN STUDY

Group	Subject #		Wt (Kg)
Control	1		94
	2		95
	3		81
	4		66
	5		79
	6		58
	7		84
		mean	80
Oat Bran	8	dropped	
	9		78
	10		72
	11		64
	12		106
	13		82
	14		70
		mean	69
RTE	15		75
	16		82
	17		77
	18	dropped	
	19		69
	20		65
	21		88
		mean	76

APPENDIX K

Weekly Weights of Subjects
(in kilograms)

SUBJECT #	WEEK							
	0	2	4-5*	5-6*	6-7*	7-8*	10	12
1	94.2	94.2	93.5	93.1	92.4	92.6	93.4	94.2
2	95.1	95.0	92.5	92.2	91.8	92.0	91.7	91.3
3	81.1	81.2	81.1	81.2	81.1	81.3	80.4	80.8
4	65.8	65.5	65.3	65.0	64.5	64.4	66.3	66.7
5	78.1	79.5	79.1	78.7	78.6	77.8	79.0	78.4
6	57.8	57.4	57.0	57.3	57.2	57.5	57.5	57.5
7	-	83.6	83.4	83.6	82.1	82.0	81.6	81.7
Mean	78.7	79.5	78.8	78.6	78.2	78.2	78.6	78.7
9	78.6	78.3	77.6	77.8	77.7	77.2	76.8	77.0
10	73.7	72.3	71.8	72.2	71.8	71.5	71.8	72.2
11	64.1	64.0	63.7	63.6	63.3	63.3	64.0	63.1
12	105.7	106.0	105.2	104.3	103.3	103.5	104.0	103.3
13	80.9	82.3	79.8	79.7	79.4	79.0	80.9	81.7
14	-	70.3	69.2	69.1	68.7	68.5	68.5	68.0
Mean	80.6	78.9	77.9	77.8	77.4	77.2	77.7	77.6
15	75.3	75.4	74.3	74.1	73.9	74.0	75.4	76.3
16	82.7	81.4	80.9	80.7	80.1	79.7	80.0	80.2
17	77.0	76.4	76.5	77.0	77.3	77.5	78.1	77.5
19	68.5	69.0	68.1	68.1	68.2	68.2	70.0	68.5
20	75.7	75.0	75.7	75.4	75.8	75.6	75.7	75.7
21	-	87.8	87.2	86.8	85.8	85.6	85.7	85.4
Mean	75.8	77.5	77.1	77.0	76.9	76.8	77.5	77.3

* Mean of 7 daily weights

APPENDIX L

Duration and Dose of Medication Taken by Subjects During Experimental Period

SUBJECT #	MEDICATION	UNIT DOSE	DURATION/DATES
1	Aspirin	6 - 9	every day
3	Extra Strength Tylenol	1	10/26
3	"	4	11/5, 11/9
3	"	2	11/12
3	Tylenol-3	2	11/6, 11/7
3	"	1	11/8, 11/9, 11/10
5	Extra Strength Tylenol	1	10/24, 11/1
5	Tylenol	2	11/13
7	Aspirin	2	10/25, 10/27, 10/31
7	"	6	11/2, 11/3
7	Excedrin	2	10/23
7	Maalox	1	10/26, 10,28, 11/11
7	"	2	
7	Novahistine	2 oz.	11/3
7	"	1 oz.	11/4
10	Sinutab	3	10/24
10	"	5	10/25
10	"	6	10/26
10	"	4	10/28
10	Alka-Seltzer	2	10/22, 10/23
12	Vitamin C	500 mg	10/19
14	Contac	1	10/31
15	Kaopectate	1 oz.	11/12
15	Mylanta	1 oz.	10/21, 10/22, 10/25, 10/26, 10/30, 11/3, 11/7
15	Tums	2	10/19, 10/22, 10/23, 11/4
16	Aspirin	2	11/9, 11/11
20	Ibuprofen	400 mg	10/20
20	"	200 mg	10/21
20	Aspirin	2	10/22
21	Ascriptin	75 mg	10/19
21	Aspirin	2	11/10

APPENDIX M

Sample Meal Card

DAY1/ GROUP 1
BREAKFAST

NAME_____DATE_____

Cream of Wheat	49.3g	_____
Sugar, Granulated	12.6g	_____
Streusel Bar	____g	_____
Corn Oil Margarine	3.9g	_____
Orange Juice	100.0g	_____
Whole Milk	122.0g	_____
Pineapple Juice	250.0g	_____
Coffee/Tea		_____
Other _____		_____
_____		_____
_____		_____

APPENDIX N

Blood Donation Consent Form

The undersigned consents to donate blood for the following study:

_____Quaker Study_____

I have received an explanation of the study.

A sample of blood, approximately 40 ml, will be taken.

All information obtained in the study will be held strictly confidential and will be used for statistical purposes only.

_____ _____

Date Signature

APPENDIX O

Daily Supplement Intake During Experimental Period: Individual Data

SUBJECT #	DAY OF EXPERIMENTAL PERIOD													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	0	0	0	1	1	1	1	1	1	1	1	1	1	1
2	0	0	1	1	1	1	1	1	1	1	2	2	2	1
3	0	0	0	0	0	1	1	1	1	1	1	1	1	1
4	0	0	0	0	0	0	0	0	0	0	0	0	0	0
5	0	0	1	1	1	0	1	1	2	2	2	2	2	3
6	0	0	0	0	1	1	1	1	1	1	1	1	1	1
7	1	1	1	1	1	1	1	1	1	1	1	2	2	2
9	0	0	0	0	0	1	2	1	0	1	0	0	1	0
10	1	2	3	3	3	3	3	3	3	3	3	3	3	3
11	0	0	0	0	0	0	0	0	0	0	0	0	0	0
12	1	1	1	1	1	1	2	2	2	2	2	2	2	2
13	4	4	6	6	7	7	6	6	6	6	6	6	6	6
14	0	0	2	1	1	1	2	2	2	2	2	2	2	2
15	0	0	1	0	0	0	1	1	1	1	1	0	1	1
16	0	0	0	0	0	0	0	1	1	1	1	1	1	0
17	0	1	4	3	3	4	3	4	3	3	4	4	4	4
19	2	2	2	3	3	4	6	5	6	6	7	6	6	6
20	1	1	2	3	3	4	5	5	5	5	5	5	6	6
21	0	0	0	1	1	2	2	2	2	1	2	2	2	2

SUBJECT #	DAY OF EXPERIMENTAL PERIOD													
	15	16	17	18	19	20	21	22	23	24	25	26	27	28
1	2	2	2	2	2	2	3	3	3	3	3	3	3	3
2	2	2	2	2	2	2	3	3	3	3	3	3	3	3
3	1	0	0	1	1	1	1	1	0	0	1	1	1	1
4	0	0	0	0	0	0	0	0	0	0	0	0	0	0
5	2	2	2	2	2	3	3	3	3	3	2	3	4	3
6	1	1	0	1	1	1	1	1	1	1	1	1	1	1
7	2	2	3	3	3	3	3	3	3	3	3	3	3	3
9	1	0	0	0	0	0	0	0	0	0	1	1	1	1
10	3	3	3	3	3	3	3	3	3	3	3	3	2	2
11	0	0	0	0	0	0	0	0	0	0	0	0	0	0
12	2	2	2	2	2	2	3	3	3	3	3	3	3	3
13	6	6	6	6	4	6	6	6	6	6	4	6	6	6
14	2	2	2	1	2	2	2	3	2	2	2	2	2	2
15	1	1	1	1	1	1	1	1	1	1	0	1	1	1
16	1	1	1	0	1	1	0	1	1	1	1	1	1	1
17	4	4	4	5	3	5	3	4	3	5	3	5	4	4
19	6	6	6	8	6	6	6	6	6	6	6	8	6	6
20	7	6	6	7	5	3	6	5	6	6	6	5	5	6
21	2	2	2	2	2	2	3	3	3	3	3	3	3	3

APPENDIX P

Daily Coffee and Tea Intake
During Experimental Period: Individual Data

SUBJECT #		DAY OF EXPERIMENTAL PERIOD													
BEVERAGE		1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	Coffee	2	3	3	5	5	4	4	6	4	4	3	2	4	3
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2	Coffee	5	0	5	5	3	4	5	3	5	4	4	5	4	4
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
3	Coffee	2	0	1	1	3	2	2	2	2	2	2	2	2	2
	Tea	1	0	0	0	0	0	0	0	0	0	0	0	0	0
4	Coffee	2	2	2	3	3	2	1	2	3	2	2	2	2	1
	Tea	0	0	0	1	0	0	1	0	0	0	0	1	2	1
5	Coffee	1	0	2	1	3	1	1	3	3	2	2	3	4	1
	Tea	0	1	0	0	0	0	0	0	1	0	1	0	0	0
6	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	1	0	0	0	0	0	0	1	0	0	0	1	0	0
7	Coffee	6	4	4	4	2	2	2	3	5	3	3	3	3	1
	Tea	0	0	0	0	1	0	0	0	0	0	0	0	2	3
9	Coffee	3	1	3	1	3	2	2	2	2	1	1	1	2	2
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
10	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
11	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	2	1	3	2	1	1	1	1	2	1	1	1	1	1
12	Coffee	2	1	2	4	3	3	3	2	4	4	4	2	4	3
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
13	Coffee	0	0	0	0	1	0	0	0	0	0	0	0	0	0
	Tea	0	2	1	3	2	3	1	0	0	0	0	0	0	0
14	Coffee	2	0	1	1	2	1	9	2	1	2	2	2	2	2
	Tea	0	1	2	2	1	0	2	1	2	0	0	2	1	1
15	Coffee	3	3	3	2	3	2	2	3	3	2	2	2	3	1
	Tea	0	0	2	2	2	0	0	0	0	1	0	1	0	0
16	Coffee	1	1	0	0	1	1	0	1	0	0	0	1	1	1
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
17	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
19	Coffee	1	0	0	7	0	2	2	4	4	4	2	2	6	4
	Tea	3	4	2	1	4	4	2	2	2	1	1	2	0	0
20	Coffee	2	2	2	3	2	1	2	2	2	1	1	1	0	1
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
21	Coffee	2	1	2	1	2	2	1	0	1	0	1	1	2	2
	Tea	0	0	0	1	0	0	0	1	2	0	0	0	0	0

APPENDIX P

Daily Coffee and Tea Intake
During Experimental Period: Individual Data
(Continued)

SUBJECT #		DAY OF EXPERIMENTAL PERIOD													
BEVERAGE		15	16	17	18	19	20	21	22	23	24	25	26	27	28
1	Coffee	3	5	4	4	5	4	5	5	4	5	2	4	4	4
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2	Coffee	1	4	5	4	6	6	6	3	7	4	7	5	6	7
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
3	Coffee	2	2	2	2	2	2	2	2	2	2	2	1	2	2
	Tea	0	0	0	0	0	0	1	0	0	0	0	0	0	0
4	Coffee	1	0	0	2	1	1	0	0	0	0	0	0	1	0
	Tea	0	2	3	1	1	2	3	3	4	3	4	4	3	3
5	Coffee	1	1	3	3	2	2	4	1	2	3	2	0	1	2
	Tea	0	0	0	0	0	1	0	0	0	0	0	0	0	0
6	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
7	Coffee	3	4	3	3	1	2	3	2	4	3	4	3	1	5
	Tea	1	0	0	1	1	0	0	0	0	0	1	0	0	0
9	Coffee	1	1	2	1	1	1	2	1	1	1	2	0	1	1
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
10	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
11	Coffee	0	0	0	0	1	0	0	0	0	0	0	1	0	0
	Tea	2	1	2	1	1	1	2	1	1	1	1	0	1	0
12	Coffee	2	3	3	2	2	3	2	2	3	2	3	2	2	2
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
13	Coffee	0	0	1	0	0	0	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
14	Coffee	1	2	1	2	3	3	3	1	2	2	2	4	4	2
	Tea	0	2	2	0	0	0	0	0	1	0	1	0	0	1
15	Coffee	2	2	2	3	3	3	2	3	3	3	4	3	3	4
	Tea	1	1	1	0	0	1	2	0	0	1	0	0	0	0
16	Coffee	0	0	1	1	0	1	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
17	Coffee	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0
19	Coffee	5	6	6	6	4	8	8	6	7	6	6	2	8	6
	Tea	0	0	0	1	1	0	0	0	0	0	0	0	0	0
20	Coffee	3	1	2	3	2	3	1	2	3	3	5	4	2	3
	Tea	1	0	0	0	0	0	0	0	0	0	0	0	0	0
21	Coffee	0	1	1	2	1	1	1	1	0	0	1	1	1	1
	Tea	0	0	0	0	0	0	0	0	0	0	0	0	0	0

APPENDIX Q

Fiber Analysis and Calculated Hemicellulose of Subjects Consuming Two Types of Oat Bran[1]

Period I

Group	Subject	mg/day NDF	mg/day ADF	mg/day Estimated Hemicellulose[2]
Control	1	36.9	17.5	19.4
	2	37.1	17.6	19.4
	4	36.4	17.0	19.4
	5	37.3	17.9	19.4
	6	36.8	17.4	19.4
	7	37.1	17.6	19.4
Mean		36.91	17.5	19.4
SEM		± 0.3	± 0.3	± 0.01
Oat Bran	10	50.4	23.7	26.7
	11	49.2	22.6	26.6
	12	50.0	23.4	26.6
	13	51.6	24.9	26.7
	14	50.0	23.4	26.6
Mean		50.2	23.6	26.6
SEM		± 0.3	± 0.3	± 0.01
RTE	15	40.2	19.4	20.8
	16	40.2	19.4	20.8
	17	41.4	20.6	20.8
	19	42.3	21.5	20.8
	20	42.0	21.2	20.8
	21	40.6	19.8	20.8
Mean		41.1	20.3	20.8
SEM		± 0.3	± 0.3	± 0.01

[1] Based on dry matter content.

[2] Estimated hemicellulose = %NDF minus %ADF

APPENDIX Q

Fiber Analysis and Calculated Hemicellulose of Subjects Consuming Two Types of Oat Bran (Continued)

Period II

Group	Subject	mg/day NDF	mg/day ADF	mg/day Estimated Hemicellulose[1]
Control	1	36.8	24.7	12.1
	2	36.8	24.7	12.1
	4	35.6	23.5	12.1
	5	36.8	24.7	12.1
	6	36.0	23.9	12.1
	7	36.8	24.7	12.1
Mean		36.5	24.4	12.1
SEM		± 0.3	± 0.3	± 0.01
Oat Bran	10	43.7	24.0	19.7
	11	42.6	22.9	19.6
	12	43.8	24.1	19.7
	13	44.9	25.2	19.7
	14	43.4	23.7	19.7
Mean		43.7	24.0	19.7
SEM		± 0.3	± 0.3	± 0.01
RTE	15	38.8	24.1	14.7
	16	38.8	24.1	14.7
	17	40.0	25.3	14.7
	19	41.0	26.3	14.7
	20	40.7	26.0	14.7
	21	39.7	25.0	14.7
Mean		39.8	25.1	14.7
SEM		± 0.3	± 0.3	± 0.01

[1] Based on dry matter content.

[2] Estimated hemicellulose = %NDF minus %ADF

APPENDIX R

Neutral Detergent Fiber Intake, Output, and Percent Digestibility, Individual Data[1]

Period I

Subject Number	Dietary Treatment	mg/day Intake[2]	mg/day Feces	% Digest.
1		36.9	7.3	80.2
2		37.1	7.9	78.8
4	Control	36.4	5.2	85.8
5		37.3	4.6	87.5
6		36.8	5.8	84.2
7		37.1	5.6	84.8
Mean		36.9	6.1	83.6
±SEM		± 0.3	± 1.7	± 3.5
10		50.4	21.6	57.1
11		49.2	6.5	86.7
12	Oat Bran	50.0	7.5	85.1
13		51.6	7.8	84.8
14		50.0	8.7	82.6
MEAN		50.2	10.4	79.3
±SEM		± 0.3	± 1.8	± 3.9
15		40.2	7.8	80.5
16		40.2	5.9	85.3
17	RTE	41.4	7.7	81.4
19		42.3	8.0	81.2
20		42.0	15.8	62.3
21		40.6	6.2	84.7
MEAN		41.1	8.6	79.2
±SEM		± 0.3	± 1.7	± 3.5

[1] Based on dry matter content.

[2] Intake represents the total amount consumed from diet and supplement.

APPENDIX R

Neutral Detergent Fiber Intake, Output and Percent Digestibility, Individual Data (Continued)

Period II

Subject Number	Dietary Treatment	mg/day Intake[1]	Feces	% Digest.
1		36.8	6.0	83.6
2		36.8	8.0	78.4
4	Control	35.6	4.3	87.8
5		36.8	6.0	83.8
6		36.0	2.8	92.3
7		36.8	4.8	87.0
Mean		36.5	5.3	85.5
±SEM		± 0.3	± 1.7	± 4.0
10		43.7	17.2	60.6
11		42.6	3.6	91.6
12	Oat Bran	43.8	8.9	79.8
13		44.9	12.2	73.1
14		43.4	19.7	54.6
MEAN		43.7	12.3	71.9
±SEM		± 0.3	± 1.9	± 4.4
15		38.8	9.1	76.6
16		38.8	5.9	84.9
17	RTE	40.0	6.6	83.6
19		41.0	14.2	65.3
20		40.7	11.7	71.3
21		39.7	5.7	85.8
MEAN		39.8	8.8	77.9
±SEM		± 0.3	± 1.7	± 4.0

[1] Intake represents the total amount consumed from diet and supplement.

APPENDIX S

Acid Detergent Fiber Intake, Output and Percent Digestibility, Individual Data[1]

Period I

Subject Number	Dietary Treatment	mg/day Intake[2]	mg/day Feces	% Digest.
1		36.9	7.3	80.2
2		37.1	7.9	78.8
4	Control	36.4	5.2	85.8
5		37.3	4.6	87.5
6		36.8	5.8	84.2
7		37.1	5.6	84.8
Mean		36.9	6.1	83.6
±SEM		± 0.3	± 1.7	± 3.5
10		50.4	21.6	57.1
11		49.2	6.5	86.7
12	Oat Bran	50.0	7.5	85.1
13		51.6	7.8	84.8
14		50.0	8.7	82.6
MEAN		50.2	10.4	79.3
±SEM		± 0.3	± 1.8	± 3.9
15		40.2	7.8	80.5
16		40.2	5.9	85.3
17	RTE	41.4	7.7	81.4
19		42.3	8.0	81.2
20		42.0	15.8	62.3
21		40.6	6.2	84.7
MEAN		41.1	8.6	79.2
±SEM		± 0.3	± 1.7	± 3.5

[1] Based on dry matter content.

[2] Intake represents the total amount consumed from diet and supplement.

APPENDIX S

Acid Detergent Fiber Intake, Output and Percent Digestibility, Individual Data (Continued)

Period II

Subject Number	Dietary Treatment	mg/day Intake[1]	Feces	% Digest.
1		36.8	6.0	83.6
2		36.8	8.0	78.4
4	Control	35.6	4.3	87.8
5		36.8	6.0	83.8
6		36.0	2.8	92.3
7		36.8	4.8	87.0
Mean		36.5	5.3	85.5
±SEM		± 0.3	± 1.7	± 4.0
10		43.7	17.2	60.6
11		42.6	3.6	91.6
12	Oat Bran	43.8	8.9	79.8
13		44.9	12.2	73.1
14		43.4	19.7	54.6
MEAN		43.7	12.3	71.9
±SEM		± 0.3	± 1.9	± 4.4
15		38.8	9.1	76.6
16		38.8	5.9	84.9
17	RTE	40.0	6.6	83.6
19		41.0	14.2	65.3
20		40.7	11.7	71.3
21		39.7	5.7	85.8
MEAN		39.8	8.8	77.9
±SEM		± 0.3	± 1.7	± 4.0

[1] Intake represents the total amount consumed from diet and supplement.

APPENDIX T

Calcium Intake, Urine and Fecal Excretion, and Retention, Individual Data[1]

Period I

Subject Number	Dietary Treatment	Intake[2]	Urine	Feces	Retention
1		1098	165	945	-13
2		1098	267	846	-14
4	Control	1097	181	695	222
5		1098	263	725	110
6		1098	352	901	-155
7		1098	182	597	319
Mean		1098	235	785	78
±SEM		± .24	± 33	± 47	± 70
10		994	73	835	86
11		993	127	770	95
12	Oat Bran	993	179	827	-14
13		994	250	885	-141
14		993	95	774	124
MEAN		993	145	818	30
±SEM		± .26	± 36	± 52	± 76
15		1136	163	996	-23
16		1136	114	728	294
17	RTE	1137	212	1055	-130
19		1138	336	1039	-236
20		1138	300	1098	-261
21		1137	115	996	26
MEAN		1137	207	985	-55
±SEM		± .24	± 33	± 47	± 70

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet, supplement, and coffee

APPENDIX T

Calcium Intake, Urine and Fecal Excretion, and Retention, Individual Data[1] (Continued)

Period II

Subject Number	Dietary Treatment	Intake[2]	Urine	Feces	Retention
1		1137	225	1030	-117
2		1137	289	950	-102
4	Control	1136	210	866	60
5		1137	264	686	187
6		1136	221	800	115
7		1137	248	951	-61
Mean		1137	243	880	14
±SEM		± .25	± 29	± 48	± 66
10		1004	142	896	-33
11		1003	148	710	145
12	Oat Bran	1004	253	854	-102
13		1005	305	948	-248
14		1004	66	823	115
MEAN		1004	183	846	-25
±SEM		± .28	± 32	± 53	± 73
15		1280	183	1136	-39
16		1280	119	998	163
17	RTE	1281	204	1240	-163
19		1282	284	1350	-352
20		1281	323	1176	-218
21		1281	171	1034	76
MEAN		1281	214	1156	-89
±SEM		± .25	± 29	± 48	± 66

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet, supplement, and coffee

APPENDIX U

Magnesium Intake, Urine and Fecal Excretion, and Retention, Individual Data[1]

Period I

Subject Number	Dietary Treatment	Intake[2]	Urine	Feces	Retention
1		239.4	66.6	188.4	-15.7
2		239.4	98.7	139.8	0.9
4	Control	239.3	82.0	129.4	27.9
5		239.4	82.3	124.2	32.9
6		239.3	131.0	185.1	-76.9
7		239.4	64.1	143.1	32.2
Mean		239.4	87.5	151.7	0.2
SEM		± .04	± 14	± 15	± 18
10		444.9	31.1	298.5	115.2
11		444.7	78.5	290.6	75.6
12	Oat Bran	444.8	60.3	355.8	28.7
13		445.0	104.5	292.3	48.2
14		444.8	69.0	294.5	81.4
MEAN		444.8	68.7	306.3	69.8
±SEM		± .04	± 15	± 17	± 19
15		427.9	63.4	374.3	-9.8
16		427.9	35.7	263.5	128.7
17	RTE	428.0	124.0	277.5	26.5
19		428.1	126.2	276.3	25.7
20		428.1	161.2	227.2	39.6
21		427.9	79.3	262.4	86.3
MEAN		428.0	98.3	280.2	49.5
±SEM		± .03	± 14	± 15	± 18

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet, supplement, coffee & tea

APPENDIX U

Magnesium Intake, Urine and Fecal Excretion, and Retention, Individual Data[1] (Continued)

Period II

Subject Number	Dietary Treatment	Intake[2]	Urine	Feces	Retention
1		281.1	77.6	202.2	1.3
2		281.1	133.2	165.5	-17.7
4	Control	280.9	82.6	156.7	41.6
5		281.1	113.6	115.6	51.9
6		281.0	85.5	149.7	45.8
7		281.1	106.0	195.9	-20.9
Mean		281.0	99.8	164.3	17.0
±SEM		± .04	± 13	± 21	± 21
10		450.2	66.8	259.9	123.6
11		450.1	99.5	275.3	75.4
12	Oat Bran	450.3	93.4	343.3	13.6
13		450.4	110.0	346.7	-5.9
14		450.2	58.6	295.4	96.2
MEAN		450.2	85.6	304.1	60.6
±SEM		± .04	± 14	± 23	± 23
15		455.9	81.5	330.2	44.2
16		455.9	44.7	416.0	-4.8
17	RTE	456.0	100.2	328.8	27.0
19		456.1	136.0	423.4	-103.3
20		456.1	172.2	253.9	30.0
21		456.0	120.0	272.2	63.9
MEAN		456.0	109.1	337.4	9.5
±SEM		± .04	± 13	± 21	± 21

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet, supplement, coffee & tea

APPENDIX V

Zinc Intake, Urine and Fecal Excretion, and Retention, Individual Data[1]

Period I

Subject Number	Dietary Treatment	Intake[2]	Urine	Feces	Retention
1		8.05	0.82	9.13	-1.90
2		8.05	0.93	8.75	-1.63
4	Control	8.05	0.63	7.21	0.21
5		8.06	0.51	6.54	1.01
6		8.05	1.18	7.33	-0.46
7		8.05	0.58	5.40	2.07
Mean		8.05	0.78	7.39	-0.12
±SEM		± .00	± .13	± .58	± .59
10		11.20	0.63	7.63	2.94
11		11.20	0.40	9.68	1.12
12	Oat Bran	11.20	0.31	10.85	0.04
13		11.21	0.64	11.23	-0.66
14		11.20	0.91	8.84	1.45
MEAN		11.20	0.58	9.65	0.98
±SEM		± .00	± .13	± .64	± .65
15		12.09	0.34	10.57	1.18
16		12.09	0.77	6.96	4.36
17	RTE	12.10	0.43	9.96	1.71
19		12.11	1.39	9.28	1.43
20		12.10	1.13	10.62	0.35
21		12.09	0.79	8.39	2.91
MEAN		12.10	0.81	9.30	1.99
±SEM		± .00	± .13	± .58	± .59

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet and supplement

APPENDIX V

Zinc Intake, Urine and Fecal Excretion, and Retention, Individual Data[1] (Continued)

Period II

Subject Number	Dietary Treatment	Intake[2]	Urine	Feces	Retention
1		9.31	0.79	9.03	-0.51
2		9.31	0.83	7.94	-0.54
4	Control	9.30	0.63	6.94	1.73
5		9.31	0.53	5.64	3.15
6		9.30	0.77	6.34	2.19
7		9.31	0.73	8.98	-0.40
Mean		9.30	0.71	7.48	1.12
±SEM		± .00	± .13	± .59	± .63
10		11.7	0.99	9.75	0.92
11		11.7	0.43	7.96	3.27
12	Oat Bran	11.7	0.32	9.99	1.36
13		11.7	1.10	12.23	-1.66
14		11.7	0.67	8.11	2.88
MEAN		11.67	0.70	9.61	1.35
±SEM		± .00	± .14	± .65	± .69
15		11.33	0.30	10.73	0.30
16		11.33	0.79	8.41	2.14
17	RTE	11.34	0.46	9.87	1.02
19		11.35	1.35	11.10	-1.10
20		11.35	0.98	10.20	0.17
21		11.34	1.23	8.18	1.93
MEAN		11.34	0.85	9.75	0.74
±SEM		± .00	± .13	± .59	± .63

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet and supplement

APPENDIX W

Copper Intake, Fecal Excretion, and Retention, Individual Data[1]

Period I

Subject Number	Dietary Treatment	Intake[2]	Feces	Retention
1		1.72	1.67	0.05
2		1.72	1.73	-0.01
4	Control	1.72	1.28	0.44
5		1.72	1.15	0.57
6		1.72	1.41	0.31
7		1.72	1.06	0.66
Mean		1.72	1.38	0.34
±SEM		± .00	± .12	± .12
10		2.08	1.42	0.66
11		2.08	2.30	-0.22
12	Oat Bran	2.08	1.49	0.59
13		2.08	1.80	0.28
14		2.08	1.38	0.70
MEAN		2.08	1.68	0.40
±SEM		± .00	± .13	± .13
15		2.20	1.92	0.28
16		2.20	1.39	0.81
17	RTE	2.20	1.67	0.53
19		2.20	1.77	0.43
20		2.20	1.88	0.32
21		2.20	1.71	0.49
MEAN		2.20	1.72	0.48
±SEM		± .00	± .12	± .12

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet and supplement

APPENDIX W

Copper Intake, Fecal Excretion, and Retention, Individual Data[1] (Continued)

Period II

Subject Number	Dietary Treatment	Intake[2]	Feces	Retention
1		1.68	1.55	0.13
2		1.68	1.52	0.16
4	Control	1.68	1.32	0.36
5		1.68	0.98	0.70
6		1.68	1.15	0.53
7		1.68	1.35	0.33
Mean		1.68	1.31	0.37
±SEM		± .00	± .14	± .14
10		2.23	1.59	0.64
11		2.23	1.21	1.02
12	Oat Bran	2.23	1.40	0.83
13		2.23	1.61	0.62
14		2.23	1.36	0.87
MEAN		2.23	1.43	0.80
±SEM		± .00	± .15	± .15
15		2.23	1.57	0.66
16		2.23	1.41	0.82
17	RTE	2.23	1.56	0.67
19		2.23	1.83	0.40
20		2.23	2.81	-0.58
21		2.23	1.61	0.62
MEAN		0.22	1.80	0.43
±SEM		± .00	± .14	± .14

[1] Reported in mg/day

[2] Intake represents the total amount consumed from diet and supplement

APPENDIX X

Plasma Calcium Values : Individual Data[1]

Subject #	Weeks 9/20	10/04	10/17	10/18	10/25	11/01	11/08	11/14	11/15	11/29	12/13
1	9.87	10.54	10.41	10.32	9.71	9.42	9.48	9.55	-	9.98	9.88
2	9.87	10.38	10.48	10.48	9.00	9.71	9.95	10.11	9.39	9.83	9.66
3	9.48	10.41	10.70	10.32	9.26	9.23	9.39	9.80	9.51	9.51	9.95
4	9.55	10.35	10.35	9.61	9.35	9.23	9.55	9.85	8.86	8.58	9.35
5	9.87	10.19	10.19	9.77	9.19	8.71	9.45	-	8.83	9.01	8.88
6	9.77	9.64	-	9.96	9.39	9.29	9.51	9.19	9.42	9.33	10.05
7	-	10.16	10.06	10.12	9.10	9.48	9.36	9.23	9.51	9.02	9.68
Mean	9.67	10.19	10.32	10.04	9.21	9.26	9.47	9.58	9.22	9.29	9.57
± SD	1.8	3.0	2.5	3.2	2.6	3.0	2.1	3.9	3.3	5.0	4.1
9	9.51	9.99	9.90	9.39	8.81	9.01	9.36	9.27	9.08	9.14	9.48
10	9.26	-	10.00	9.87	9.61	9.26	-	9.05	8.80	8.92	9.28
11	9.67	10.09	10.06	9.93	9.26	8.74	9.42	8.71	8.84	9.17	9.48
12	9.06	9.22	9.67	9.16	8.94	8.74	-	-	8.92	-	9.05
13	9.87	9.71	10.38	9.71	9.39	9.61	9.70	-	9.55	9.70	9.05
14	-	10.96	10.73	10.32	9.64	9.71	9.76	-	9.48	10.14	9.61
Mean	9.42	9.94	10.05	9.68	9.23	9.15	9.53	8.97	9.05	9.38	9.27
± SD	3.2	6.2	3.9	4.3	3.4	4.3	2.1	2.5	3.3	5.0	2.4
15	9.80	9.38	10.48	10.41	9.39	9.58	9.92	10.01	9.59	9.61	10.01
16	9.90	10.99	10.64	10.35	9.39	9.93	9.80	9.92	9.55	9.61	9.91
17	9.64	10.32	10.22	9.55	9.16	9.45	-	9.36	9.39	9.23	9.01
19	9.77	9.77	10.64	10.06	9.71	9.80	9.51	9.80	9.45	9.67	10.11
20	9.74	10.41	10.19	10.06	9.42	9.61	9.61	-	9.48	9.58	10.05
21	-	9.93	10.93	10.00	9.39	9.55	9.12	9.58	9.48	9.23	9.91
Mean	9.72	10.08	10.47	10.02	9.35	9.62	9.56	9.68	9.42	9.45	9.82
± SD	0.8	5.7	2.9	3.2	2.0	1.9	3.0	2.9	0.8	2.0	4.1

[1] Reported as mg/dl.

APPENDIX Y

Plasma Magnesium Values : Individual Data[1]

Subject #	Weeks 9/20	10/04	10/17	10/18	10/25	11/01	11/08	11/14	11/15	11/29	12/13
1	1.72	1.72	1.75	1.82	1.57	1.63	1.67	1.68	-	1.77	1.70
2	1.67	1.75	1.72	1.84	1.44	1.62	1.69	1.64	1.69	1.58	1.58
3	1.73	1.68	1.70	1.75	1.53	1.66	1.65	1.63	1.80	1.63	1.69
4	1.75	1.82	2.00	2.01	1.73	1.75	1.83	1.68	1.79	1.75	1.91
5	1.71	1.68	1.82	1.79	1.57	1.62	1.70	-	1.74	1.57	1.65
6	1.72	1.65	-	1.81	1.65	1.66	1.85	1.57	1.74	1.83	1.88
7	-	1.82	1.77	1.74	1.67	1.76	1.86	1.52	1.78	1.70	1.74
Mean	1.72	1.73	1.79	1.82	1.59	1.67	1.75	1.67	1.76	1.69	1.74
± SD	0.3	0.7	1.1	0.9	1.0	0.6	0.9	0.8	0.4	1.0	1.2
9	1.79	1.74	1.85	1.74	1.62	1.74	1.84	1.86	1.82	1.92	1.84
10	1.51	-	1.66	1.76	1.55	-	-	1.75	1.73	1.71	1.62
11	1.63	1.67	1.66	1.68	1.60	1.62	1.68	1.56	1.56	1.72	1.68
12	1.86	1.68	1.91	1.88	1.77	1.81	-	-	1.87	-	1.82
13	1.75	1.64	1.89	1.76	1.73	1.72	1.81	-	1.86	1.80	1.65
14	-	1.88	1.86	1.98	1.69	1.68	1.86	-	1.90	1.89	1.91
Mean	1.71	1.72	1.81	1.80	1.66	1.71	1.80	1.72	1.79	1.81	1.75
± SD	1.4	1.0	1.1	1.1	0.9	0.7	0.8	1.5	1.3	0.9	1.2
15	1.72	1.59	2.00	1.75	1.75	1.73	1.80	1.92	1.74	1.65	1.90
16	2.01	1.92	1.95	1.96	1.84	1.87	1.90	2.06	2.05	1.95	1.88
17	1.72	1.80	1.88	1.80	1.62	1.68	-	1.77	1.77	1.63	1.63
19	1.58	1.63	1.74	1.73	1.61	1.62	1.62	1.77	1.68	1.60	1.67
20	1.66	1.82	1.69	1.84	1.63	1.64	1.79	-	1.79	1.65	1.82
21	-	1.72	1.77	1.84	1.71	1.74	1.63	1.78	1.77	1.82	1.79
Mean	1.74	1.75	1.84	1.82	1.69	1.71	1.75	1.86	1.80	1.72	1.78
± SD	1.6	1.2	1.2	0.8	1.0	0.9	1.2	1.3	1.3	1.4	1.1

[1] Reported as mg/dl.

APPENDIX Z

Plasma Zinc Values : Individual Data[1]

Subject #	Weeks 9/20	10/04	10/17	10/18	10/25	11/01	11/08	11/14	11/15	11/29	12/13
1	105.6	120.7	120.9	118.0	110.7	109.2	109.2	117.2	-	103.3	114.4
2	114.7	98.1	141.5	125.4	127.9	106.3	101.9	112.5	104.8	-	116.0
3	96.6	81.5	93.1	107.8	123.1	104.8	119.1	117.2	100.2	103.3	112.8
4	101.1	69.3	113.6	99.0	106.5	96.1	97.5	97.1	94.2	78.5	103.3
5	92.0	84.5	125.3	100.5	97.5	85.9	90.2	-	84.7	89.3	103.3
6	105.6	89.0	-	99.0	106.3	97.5	103.4	101.7	101.7	100.1	109.7
7	-	84.5	112.2	108.1	100.5	97.5	100.5	107.9	106.4	97.3	120.3
Mean	102.6	89.7	117.8	108.3	110.4	99.6	103.1	108.9	98.7	95.3	111.4
± SD	7.9	16.2	16.0	10.2	11.3	7.9	9.1	8.3	8.0	9.7	6.4
9	101.1	97.0	119.5	101.7	103.4	103.4	106.3	115.6	104.8	90.9	106.3
10	84.5	-	106.3	99.0	-	-	-	100.2	-	70.7	-
11	98.1	92.0	101.9	97.5	97.5	96.1	99.0	104.8	97.3	86.2	104.8
12	82.9	90.7	101.9	84.3	87.3	87.3	-	-	94.2	-	94.0
13	93.8	72.4	109.2	96.1	94.6	111.2	100.5	-	98.6	108.1	101.7
14	-	86.0	123.9	104.8	100.5	99.0	85.8	-	94.2	106.5	94.0
Mean	92.1	87.6	110.5	97.2	96.7	99.4	97.9	106.9	97.8	92.5	100.2
± SD	8.1	9.4	9.2	7.1	6.2	8.8	8.7	7.9	4.4	15.5	5.9
15	104.1	92.0	126.8	109.2	107.8	107.8	106.3	115.6	109.5	119.1	109.4
16	119.2	96.6	112.2	110.7	106.5	107.8	99.0	107.9	103.2	109.7	112.8
17	122.3	98.1	119.5	90.1	97.5	100.5	-	107.9	98.6	-	100.1
19	115.9	104.1	137.0	119.1	106.3	118.0	107.8	121.8	109.5	109.7	121.8
20	117.7	96.6	123.9	118.0	106.3	-	102.0	-	101.7	114.4	124.9
21	-	130.2	122.4	109.2	103.4	97.5	97.5	117.2	106.4	122.3	106.3
Mean	115.8	102.9	123.6	109.4	104.6	106.3	102.5	114.1	104.8	115.0	112.6
± SD	7.0	13.9	8.2	10.4	3.8	7.9	4.5	6.1	4.4	5.6	9.4

[1] Reported as μg/dl.

VITA

Susan H. Vyule (formly Susan Kay Hiner) was born on May 13, 1960 and lived in Highland County, Virginia where she attended Highland County High School. She graduated salutatorian of the Class of 1978. In the fall of 1978 she entered the College of Home Economics at Virginia Polytechnic Institute and State University from which she received a B.S. degree in Human Nutrition and Foods with a minor in Sociology in the spring of 1982. During 1982, she enrolled in the graduate program at Virginia Polytechnic Institute and State University in Human Nutrition and Foods to pursue a Master of Science degree. In April, 1984 she married Benjamin H. Vyule and left the university to work full time. She returned to the university in 1993 to complete her degree. Susan has one son, Stuart.

Susan H. Vyule

Susan H. Vyule