

Nutritional Properties of Hard White and Hard Red Winter Wheats and Oatmeal. II. Effects on Fecal Water-Holding Capacity and Loss of Protein, Ash, Calcium, and Zinc in Cholesterol-Fed Rats¹

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ABSTRACT

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The effects of oatmeal and various fractions from a hard white and hard red winter wheat on rat weight gains and fecal loss of protein, ash, calcium, and zinc were compared. Oatmeal diets produced weight gains similar to those of the whole wheat flour diets. No significant differences occurred in weight gains between animals fed diets containing whole flour, straight-grade flour, or bran of the hard white wheat and those fed respective hard red winter wheat diets. Although caloric contents of the wheat bran diets were lower than those of the other diets and bran-fed rats lost more protein and ash in feces, animals fed the bran diets gained as much weight as those fed straight-grade flour diets. Feed intake was highest in bran-fed animals, and the bran diets were the least efficient feeds. Animals fed bran diets had the highest wet and dry fecal weight and highest fecal water-holding capacity; no differences occurred between the hard white and hard red winter bran diets. Fecal water-holding capacity was strongly correlated with total dietary fiber ($r = 0.9738$, $P = 0.0001$). Animals fed oatmeal lost the least amount of calcium

per day, had the lowest percent loss of calcium, and absorbed more calcium than those fed red or white wheat diets or the control diet, which contained cellulose. Greatest daily loss of zinc occurred with the wheat bran diets, but zinc concentration and daily amounts of zinc absorbed also were highest with those diets. More zinc was absorbed per day from the oatmeal diet than from all other diets, except those containing wheat brans. An inverse relationship was observed between percent calcium loss and soluble dietary fiber ($r = -0.4996$, $P = 0.003$), whereas no significant correlation occurred between insoluble and total dietary fiber or phytic acid and percent calcium loss. However, significant correlations were observed between the daily amount of zinc in feces and insoluble dietary fiber ($r = 0.8709$, $P = 0.0001$), total dietary fiber ($r = 0.9137$, $P = 0.0001$), and phytic acid ($r = 0.9552$, $P = 0.0001$). The amounts of calcium and zinc absorbed daily were strongly correlated with intake ($r = 0.9124$, $P = 0.0001$ and $r = 0.9134$, $P = 0.0001$, respectively).

Much controversy now surrounds the role of dietary fiber in bioavailability of minerals and other nutrients. Interest in the availability of essential minerals in the diet has increased because of studies indicating that diets with high levels of fiber impair mineral utilization (Gordon 1990). Some investigators have reported that dietary fiber binds minerals in the digestive tract and renders them unavailable for absorption (Mod et al 1985). Others report that phytate is the major determinant of mineral availability, with fiber being of little or no significance. Some evidence suggests that both fiber and phytate inhibit utilization of minerals (Gordon 1990).

Mechanisms by which dietary fiber influences mineral absorption are related to its physicochemical properties (Kay 1982). Those properties include the ability of dietary fiber to: 1) act as a weak cation exchanger, 2) shorten intestinal transit time, 3) dilute mineral concentration by increasing fecal bulk, and 4) resist digestion in the large bowel. Toma and Curtis (1986) suggested that fiber might interfere with formation of a calcium-binding protein complex, an essential factor for the transport of calcium within the intestine.

In vitro experiments indicate that wheat bran and its components have little affinity for binding calcium (Rendleman 1982, Rendleman and Grobe 1982). However, Dintzis et al (1985) found that humans consuming 26 g per day of wheat bran, corn bran, or soybean hulls had increased fecal excretion of calcium.

Most animal studies indicate that dietary fiber alone does not have a major effect on zinc availability. Wheat bran at levels of up to 15% in the diet did not significantly affect zinc absorption in rats (Borgheri 1982). That was confirmed by Caprez and Fairweather-Tait (1982) using milled wheat bran.

Studies to determine the effects of wheat fractions on mineral absorption often have not specified the type of wheat used or adequately described the fraction being tested. Many studies have

reported the effects of oatmeal on serum cholesterol levels, but few included effects on mineral loss.

The objective of this experiment was to determine the effects of hard white and hard red winter wheat and their fractions and oatmeal on fecal nutrient loss and water-holding capacity (WHC) in rats. That was accomplished using appropriate statistical analyses and comparisons to determine significant relationships among dietary factors and nutrient loss.

MATERIALS AND METHODS

Materials

Hard red winter wheat [Norkan (88-854)] and hard white winter wheat [KS84HW196 (88-850)] cultivars grown in 1988 at Fort Hays Branch Experiment Station, Hays, KS were obtained. Each wheat cultivar was milled to whole wheat flour (100% of the grain, Europemill EM-25/251, Denmark) or straight-grade flour of 74% extraction and bran (26% of the grain) using the Miag Multomat S/100 (Braunschweig, Germany). After milling, fractions were stored in a cold room (2°C) until needed. Bran particle size was not reduced before addition to the rat diets. Diets were mixed in a Wenger ribbon mixer (Wenger Manufacturing Co., Sabetha, KS).

Rat Feeding Study

Eight groups of 10 male Wistar rats (Charles River Laboratories, Wilmington, MA) were fed diets containing the following for eight weeks: 1) casein-based diets with alphacel cellulose (control); 2) hard white wheat whole flour; 3) hard red wheat whole flour; 4) hard white wheat bran; 5) hard red wheat bran; 6) hard white wheat straight-grade flour; 7) hard red wheat straight-grade flour; or 8) oatmeal (Table I).

Diets were formulated to contain 20% protein and 6% fat and were the same as those used in Part I of this study (Maziya-Dixon and Klopfenstein 1994), except that wheat bran diets contained 50% instead of 45% bran, with other ingredients adjusted accordingly (Table I). In addition, the wheat bran was not hammermilled (1.5-mm screen) before addition to the diets as it was in Part I. Standard AACC (1983) methods were used in

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analyzing diets for ash (method 08-01); insoluble dietary fiber (IF), soluble dietary fiber (SF), and total dietary fiber (TDF) (method 32-07) (Table II). Wheat bran diets were high in TDF and IF. The oatmeal diet had about equal amounts of SF and IF. TDF was lowest for the straight-grade flour diets, and whole wheat flour diets had intermediate TDF levels.

Animals, initially weighing 129 ± 5 g, were individually housed in stainless steel cages in an environmentally controlled room with a 12-hr light-dark cycle. Diets were provided ad libitum. Animals were weighed weekly, feed consumption records were kept, and feed efficiencies were calculated.

Phytic Acid

The method of Tangkongchitr et al (1981) was used to determine phytic acid content of wheat fractions (whole flours, brans, and straight-grade flours) and oatmeal. Phytic acid was determined colorimetrically by the method of Lindberg and Ernster (1956) as modified by Nahapetian and Bassiri (1975), and content in the diets was calculated (Table II).

The bran diets had the highest concentrations of phytic acid. With the exception of the control diet, the straight-grade flour diets had the lowest concentrations of phytic acid, and the whole flour and oatmeal diets were intermediate between the bran and straight-grade flour diets. The red and white wheats showed no significant differences in phytic acid concentration. Phytic acid in the grains was highly correlated with IF ($r = 0.9579$, $P = 0.0001$) and TDF ($r = 0.9819$, $P = 0.0001$).

Mineral Analyses and Fecal WHC

Calcium and zinc of rat diets and feces were determined by atomic absorption spectroscopy (method 40-70, AACC 1983) after dry ashing. Feces for mineral analyses and WHC determination were collected daily during weeks 5 and 6 (total of 14 days) of the feeding study. At each collection, feces were placed in styro-foam cups with close-fitting lids, then frozen until the next collection. Although some moisture was lost during the collection period, losses were minimized by frequent collection and cold storage in sealed containers. Moisture content of feces was deter-

mined by drying to a constant weight in an air oven at 50°C . The relatively low drying temperature was used to minimize heat damage to biochemical components of the feces (which were to be assayed later), while preventing the growth of the mesophilic bacteria normally present in feces (Wilson et al 1979). WHC was calculated as: $100 \times (\text{feces wet weight} - \text{feces dry weight}) / \text{feces dry weight}$.

All diets met the rats' dietary requirement for calcium (0.05 g/100 g) and zinc (0.012 mg/g) (NRC 1978). Among the different wheat fractions, the brans had the highest concentrations of both calcium and zinc (Table II), and the straight-grade flour diets had the lowest concentrations. Calcium and zinc concentrations were higher in the oatmeal diet than in the whole wheat diets. Fecal protein and ash were measured using the same methods as for the diet analyses.

Statistical Procedures

Data were analyzed by the Statistical Analysis System (SAS 1989) at Kansas State University, using one-way analysis of variance with the Fisher's protected least significant difference (LSD) test (Ott 1988) and correlation analysis (Pearson) by group by the Proc Corr procedure. A complete randomized experimental design was used.

RESULTS AND DISCUSSION

Overall Weight Gains, Feed Efficiencies, and Feed Intake

No significant differences occurred in weight gains between animals fed respective fractions of hard white versus hard red winter wheat, but the red whole flour and red straight-grade flour diets were more efficient feeds than the respective white wheat diets (Table III). Feed efficiencies were lower for all diets than they were in Part I of this study (Maziya-Dixon and Klopfenstein 1994) in which initial weights of the rats were higher and their digestive systems presumably were more mature and better able to utilize the diets. Animals fed oatmeal had weight gains similar to those of animals fed red or white whole wheat. Those fed the control diet gained the least weight, but not significantly less

TABLE I
Percent Composition of Rat Diets Containing Wheat, Oatmeal, or Cellulose^a

Diet	Content	Cereal	Casein ^b	Corn Starch ^b	Fat ^c	Cellulose ^d
1	Control	0	23.5	47.0	5.0	6.0
2	White whole flour	50	14.9	12.4	4.2	...
3	Red whole flour	50	13.4	13.9	4.1	...
4	White bran	50	12.1	16.3	3.1	...
5	Red bran	50	11.2	17.1	3.2	...
6	White straight-grade flour	50	14.7	12.1	4.6	...
7	Red straight-grade flour	50	14.9	12.0	4.5	...
8	Oatmeal	50	13.0	17.1	1.4	...

^aAll diets contained 4% salt mixture XVII, 1% vitamin mix 2 (both obtained from ICN Nutritional Biochemicals, Cleveland, OH), 1% cholesterol, 0.30% DL-methionine, 0.2% choline bitartrate (all from Sigma Chemical Co., St. Louis, MO), and 12% sucrose.

^bVitamin-free casein and corn starch from Sigma.

^cVegetable oil (soybean) from a local supermarket.

^dAlphacel from ICN.

TABLE II
Ash, Dietary Fiber, Phytic Acid, Calcium, Zinc, and Caloric Content of Rat Diets (as Fed)^a

Diet	Content	Ash (%)	IF (%)	SF (%)	TDF (%)	Phytic Acid (%)	Calcium (mg/g)	Zinc (mg/g)	Energy (kcal/100 g)
1	Control	3.21	4.75	0.00	4.75	0.01	2.50	0.140	384
2	White whole flour	5.64	5.90	1.56	7.46	0.48	4.60	0.250	393
3	Red whole flour	5.90	5.82	1.44	7.26	0.45	4.50	0.290	393
4	White bran	6.12	17.96	2.84	20.80	1.70	5.20	0.640	317
5	Red bran	6.60	19.29	3.43	22.72	1.76	5.45	0.590	317
6	White straight-grade flour	4.01	0.61	1.02	1.63	0.09	3.60	0.190	406
7	Red straight-grade flour	4.60	0.65	0.97	1.62	0.08	3.20	0.170	406
8	Oatmeal	6.42	3.73	3.37	7.10	0.59	4.90	0.370	390

^aIF = insoluble dietary fiber, SF = soluble dietary fiber, TDF = total dietary fiber.

than those fed the wheat bran diets. The oatmeal diet was a more efficient feed than the white wheat diets. Animals fed the straight-grade flour diets consumed the same amount of feed as those fed control and oatmeal diets but less feed than animals fed whole flour or bran diets.

Among the different wheat fractions, bran was the least efficient feed. In general, as TDF content of diets increased, feed intake increased ($r = 0.5820$, $P = 0.0001$) and feed efficiencies decreased ($r = -0.5882$, $P = 0.0001$), but the relationship between total weight gain and TDF was weak and nonsignificant.

Effects of Diets on Fecal Protein and Ash

No significant differences were observed in the daily amount of protein lost in feces of animals fed white versus red brans or whole flours (Table III), but animals fed the white straight-grade flour diet excreted a higher daily amount of protein than those fed red straight-grade flour diet. Animals fed either of the brans excreted more protein than animals fed the whole or straight-grade flours or oatmeal, and those fed a diet containing a cereal product excreted more protein than animals fed the control diet without cereal.

No significant differences were observed in the daily amount of ash lost in feces of animals fed white versus red wheat diets (Table III). Animals fed the bran diets excreted higher daily amounts of ash than those fed the whole or straight-grade flours. All animals fed cereal products lost greater amounts of ash per day than the control animals.

Effects of Diets on Fecal WHC

Animals fed bran diets had the highest wet and dry fecal weights and highest fecal WHC with no differences between the hard white and hard red winter wheat bran diets (Table III). Animals fed oatmeal, control (cellulose), and red or white whole wheat diets had similar and intermediate wet and dry fecal weights, whereas animals fed straight-grade flour diets had the lowest wet and dry fecal weights. The only significant differences in fecal characteristics resulting from feeding white versus red wheat fractions were for the whole wheat diets. Fecal dry weight was higher and WHC was lower for animals fed red whole flour than they were for animals fed white whole flour. Fecal WHC was strongly correlated with IF ($r = 0.9738$, $P = 0.0001$) and TDF ($r = 0.9705$, $P = 0.0001$) but less strongly with SF ($r = 0.6007$, $P = 0.0001$).

High WHC and fiber were, no doubt, mainly responsible for the higher fecal wet weights with the bran diet. However, high

daily excretion of protein and ash also occurred in animals fed that diet.

Effects on Calcium Absorption

The oatmeal diet resulted in the lowest fecal calcium loss per day, the lowest percent calcium loss, and the highest daily calcium absorption. Animals fed the control diet had the highest percent calcium loss and absorbed less dietary calcium than those fed any cereal-containing diet. A high fecal calcium loss has been reported previously in cellulose-fed animals (Klopfenstein 1990). Animals fed whole wheat diets absorbed more calcium than those fed straight-grade flour diets or red bran.

The daily amount of calcium absorbed was strongly correlated with dietary calcium intake ($r = 0.9124$, $P = 0.0001$). An inverse relationship was observed between percent calcium lost in feces and SF ($r = -0.4996$, $P = 0.0003$), whereas there was no significant correlation between IF and TDF or phytic acid and percent calcium loss. Fermentation of soluble fibers in the colon may result in lowered pH values. That could make the calcium more soluble, thereby enhancing its absorption (Clydesdale 1989).

Effects on Zinc Absorption

Although bran-fed animals lost more fecal zinc per day than any other group, actual amount absorbed was highest in those animals. Rats fed oatmeal absorbed as much zinc per day as those fed whole wheat flours. Although percent zinc loss was higher in all cereal-fed animals than in control animals, no group fed 50% cereal product diets absorbed significantly less zinc than control animals. As was noted for calcium, there was a strong positive relationship between dietary zinc intake and daily amount of zinc absorbed ($r = 0.9134$, $P = 0.0001$).

The ability of phytic acid to impair mineral bioavailability has long been known (Clydesdale 1989). Erdman (1981) suggested that zinc forms an insoluble complex with phytate, causing higher fecal loss. Dietary phytase activity varies in different grains and should also be an important factor in mineral bioavailability. In the present experiment, phytate concentration was much higher in the bran diets (>1.5%) than it was in the oatmeal diet (0.59%), but phytase activity in the wheat diets was probably higher also (Moak et al 1987). In rats, dephytinized bran resulted in significantly higher apparent zinc retention than from whole bran (Erdman 1981, Caprez and Fairweather-Tait 1982). In addition, Gordon (1990) reported that excess dietary zinc overcame the inhibitory effect of phytic acid on absorption of zinc. The present study showed significant relationships between the daily amount

TABLE III
Effects of Hard Red and Hard White Wheats or Oatmeal Diets in Rats after Six Weeks of Feeding^a

Measurement	Diet							
	Control	White Whole Flour	Red Whole Flour	White Bran	Red Bran	White Straight-Grade Flour	Red Straight-Grade Flour	Oatmeal
Weight gain (g)	165 c	192 ab	208 a	181 b	176 bc	171 bc	193 ab	214 a
Feed efficiency ^b	0.225 bc	0.238 b	0.257 a	0.210 cd	0.202 d	0.234 b	0.265 a	0.269 a
Feed intake (g)	731 cd	802 a-c	808 ab	857 ab	870 a	729 d	727 d	793 b-d
Fecal protein (mg/day)	5.8 d	8.5 bc	9.5 b	14.8 a	15.4 a	8.7 b	7.4 c	9.5 b
Fecal ash (mg/day)	134 d	234 c	276 bc	438 a	487 a	264 bc	239 c	291 b
Fecal wet weight (g/2 weeks)	22.2 c	23.4 bc	27.0 b	66.6 a	65.2 a	15.5 d	13.9 d	22.7 c
Fecal dry weight (g/2 weeks)	17.2 c	15.9 c	19.9 b	30.9 a	31.2 a	12.4 d	11.2 d	17.2 c
Water holding capacity ^c	28.7 cd	48.6 b	35.6 c	115.5 a	111.2 a	25.5 cd	24.7 d	32.6 cd
Fecal calcium loss (mg/day)	29.5 bc	31.4 bc	30.0 bc	38.2 ab	40.4 a	25.9 cd	24.2 cd	17.2 d
Percent calcium loss ^d	68.4 a	30.3 cd	23.6 de	39.6 bc	45.5 b	35.6 bc	41.5 bc	16.3 e
Calcium absorbed (mg/day) ^e	14.3 e	65.6 b	81.8 a	61.8 b	47.1 c	43.3 cd	36.0 d	90.2
Fecal zinc loss (mg/day)	1.52 e	3.97 c	5.76 b	9.47 a	9.97 a	3.13 d	2.90 d	5.75 b
Percent zinc loss ^d	61.3 c	76.4 b	95.1 a	69.5 bc	72.1 b	87.3 a	91.6 a	72.4 b
Zinc absorbed (mg/day) ^e	0.98 cd	1.24 c	0.32 d	4.46 a	3.83 a	0.46 d	0.33 d	2.27 b

^a Means in the same row not followed by the same letter are significantly different at $P < 0.05$.

^b Grams gained/gram feed consumed.

^c $100 \times (\text{feces wet weight} - \text{feces dry weight}) / \text{feces dry weight}$.

^d $100 \times (\text{weight in feces}) / \text{weight consumed}$.

^e Mg intake/day - mg fecal loss/day.

of zinc in feces and IF ($r = 0.8709$, $P = 0.0001$), TDF ($r = 0.9137$, $P = 0.0001$), and phytic acid ($r = 0.9552$, $P = 0.0001$), but also a strong correlation with dietary zinc ($r = 0.9580$, $P = 0.0001$).

CONCLUSIONS

Animal responses to oatmeal or red or white wheat whole flour diets were similar with regard to: weight gain; feed efficiency; feed intake; fecal protein and ash; and fecal wet and dry weights and WHC. However, significantly more zinc was absorbed from the oatmeal diet than from either the red or white whole flour diets. Effects of the red and white wheat bran diets were the same, except that more calcium was absorbed per day from the white bran diet. Animals consuming 50% wheat bran in their diets gained as much weight as those consuming a casein-based diet containing 6% cellulose, although fecal nutrient loss was greatest in bran-fed animals.

Much has been written about the possible negative effects of fiber-rich products on mineral and other nutrient loss in animals and humans. However, this experiment showed that percent calcium loss was much lower and more calcium was absorbed by animals consuming the phytate-rich wheat products or oatmeal than those consuming the casein-cellulose control diet. That was true even in the case of animals fed both of the straight-grade flour diets, whose dietary calcium level was similar to that of control animals. Percent calcium loss in the feces was inversely correlated with dietary SF, but not statistically related to TDF, IF, or phytic acid.

Conversely, percent zinc lost in the feces was higher in animals fed the straight-grade flour diets than in controls, even though zinc levels were similar in control and flour diets. Daily zinc loss was highly correlated with TDF, IF, and phytic acid, but not with SF. It is important to note that although daily and percent loss of zinc was higher in all grain-fed animals than the control, daily zinc absorption was not significantly lower in any cereal-fed group than in the control group. In calcium- and zinc-replete animals, daily amounts of calcium and zinc absorbed were as strongly related to concentration of those minerals in the diets as to any other dietary factor.

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