

Effect of Increasing Levels of Hard Wheat Fiber on Fecal Weight, Minerals and Steroids and Gastrointestinal Transit Time in Healthy Young Women¹

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ABSTRACT Hard red wheat bran (HRWB) baked in a yeast-leavened bread was fed to 36 healthy young college women consuming a basal diet of traditional foods, which contained 15 ± 3 g/d dietary fiber (DF). Three levels of HRWB were added supplying, respectively, 5.7, 17.1 and 28.5 g/d DF; an additional treatment group did not receive any HRWB. Fecal collections were carried out in the last 5 d of treatment. Fecal wet weight, fecal dry weight and fecal ash increased significantly for each increase in HRWB (*P* < 0.05). Fecal dry matter percent changed significantly only at the highest level of HRWB (*P* < 0.05). After accounting for the minerals in the HRWB, there was an increased fecal loss of Ca, but not of Zn, Cu, Fe or Mg compared to the women fed no HRWB. HRWB at a level of 17.2 g/d induced faster transit times (TT) than no HRWB and 66 g/d HRWB induced faster TT than either 17.2 or 39.6 g/d HRWB (*P* < 0.05). Total daily fecal steroids were not altered by changes in HRWB. Daily total bile acid excretion increased significantly (*P* < 0.05) at the two higher levels of HRWB due primarily to higher excretion of chenodeoxycholic acid. *J. Nutr.* 116: 778-785, 1986.

INDEXING KEY WORDS dietary fiber • transit time • fecal minerals • fecal steroids • wheat fiber • hard wheat bran

It has been shown that wheat and related fibers are some of the most effective in increasing fecal weight and decreasing mouth-to-anus transit time, when added to the typical diets of industrialized societies (1, 2).

Most studies have focused on comparing the effect of one level of wheat fiber to that of similar levels of other plant fibers or to that of fiber-free diets (1). Some studies have fed subjects highly controlled diets, thus eliminating the possible intrinsic effect of fiber on consumption of other foods. Dose-response studies with graded levels of wheat fiber appear to be needed to enlarge the data base available on the effects of wheat

fiber on gastrointestinal function, fecal weight and composition in the context of a varied and normal diet, controlled only for dietary fiber intake (3-7). This expanded data base should also be valuable for a better understanding of what are the most desirable levels of dietary fiber intake (4, 8-10).

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The purpose of this study was to determine the effect of three levels of a well-defined wheat bran and a diet free from wheat bran on fecal weight and moisture, minerals, steroids and on mouth-to-anus transit time. The choice of women students living in college dormitories and consuming all their meals in the same college cafeteria allowed careful control of the baseline fiber intake, a fairly homogeneous dietary pattern and yet freedom from the monotony and artificiality of rigidly controlled diets.

MATERIALS AND METHODS

Subjects. Thirty-six healthy, young college women, 18–32 yr of age (mean age 19.8 yr), residents of Mills College, Oakland, CA, participated in the study. Written, informed consent was obtained in private from each volunteer and the protocol was approved by the College Human Studies Committee.

Diets. The baseline diet was similar for all subjects, for all periods. Consumption of breakfast, lunch and dinner in the college residence hall (Founders Commons) was a prerequisite for participation. All meals were planned and supervised by a dietitian. A daily diet record was kept by all subjects and reviewed daily by a nutritionist. A monitor was present at all meals and was responsible for giving the treatments to each subject and ensuring compliance.

The typical diet consumed in the college cafeteria during this study contained approximately 35% kcal from fat, 48% kcal from carbohydrates and 17% kcal from protein.

The menus were examined daily and in advance by a study nutritionist, who would then post them in the dining room marking: 1) foods permitted ad libitum, all food free from dietary fiber (DF) (DF-free foods), 2) foods never to be consumed (such as beans and whole grain breads) and, 3) foods that had to be consumed in controlled amounts (selected DF-containing foods, typically fruits and vegetables). All subjects had to consume four portions per day of the DF-containing foods. To further ensure sufficient baseline intake of a variety of fiber, a fiber wafer supplement (4 tablets/d) based on corn bran (2.4 g/d DF) was given to the

subjects. In addition, the white flour used in preparing the treatment breads supplied a small amount of DF. The sum of the baseline DF (DF-controlled foods, fiber wafers and white flour portion of bread treatments) supplied a total of 15 ± 3 g/d DF.

Treatments and experimental design. Hard, red wheat bran (HRWB) supplied by the American Association of Cereal Chemists (St. Paul, MN) was fed at three levels to supply graded levels of wheat fiber. Table 1 gives the pertinent analytical data for the HRWB. Two different methods of analysis (11, 12) resulted in similar DF values for the HRWB, and we have used 44% DF in HRWB for all calculations. The controlled amount of fiber present in the basal diet was used as the fourth level of fiber (HRWB-free). The bran was incorporated in a baker's yeast-fermented loaf formulated to supply a desired amount of HRWB in each slice. This bread was baked in the Mills College bakery under the supervision of one of the investigators. A white bread was baked at the same time to supply all of the same ingredients per slice as the bran bread except for the HRWB. Subjects always consumed a total of 5 slices of bread per day, including 0, 1, 3 or 5 slices of the HRWB bread, which supplied, respectively, 0, 13.2, 39.6 and 66.0 g HRWB, equivalent to 0, 5.8, 17.4 and 29.0 g DF from the bran. By consuming the same number of bread slices, there was no change in the consumption of nutrients from the white flour portion.

Table 2 shows the incomplete block design used in the study. Each subject was randomly assigned to two of the four possible levels of fiber. Each period was 13 d long, and was planned to allow fecal collections on week days only. Each subject consumed the assigned treatment for the entire period, starting on a Sunday. Week 1 was designed as adaptation to the assigned level of DF. Fecal collections began at 1500 on d 8 and continued through the following Friday at 1500. Following a weekend free from restrictions, subjects started on the second DF level, following the same adaptation and collection cycle. Transit times (TT) were measured according to Hinton (13) by giving 40 radioopaque pellets at the beginning of each fecal collection cycle, i.e. at 1500 on Sunday. Excretion of 80% of the pellets was

TABLE 1
Analysis of AACC certified hard red
wheat bran (HRWB)¹

Assay	Hard red wheat bran
Crude fiber, %	10.30
Neutral detergent residue ²	43.9
Dietary fiber ³	44.0
Lignin, %	4.10
Pectin, %	11.00
Pentosans, %	26.00
Minerals, mg/100 g	
Calcium	57.20
Copper	1.55
Iron	16.50
Magnesium	600.00
Zinc	11.50

¹American Association of Cereal Chemists, Minneapolis, MN. ²Dietary fiber analyzed by the NDR method (11). ³Dietary fiber analyzed by the method of Prosky et al. (12).

taken as the TT.

Fecal collections and analyses. Feces were collected in special containers (Commode Specimen System, Sage Products, Elk Grove, IL) and brought daily, or as often as needed, to a collection center where refrigerated facilities were available. All subjects were instructed to keep fecal containers cold until they were brought to the collection area. Styrofoam containers were supplied to the subjects for use when other methods of refrigeration were not available. The collection center was within easy walking distance of the residence halls and classroom, thus ensuring consistent handling of the fecal samples. The feces were then weighed for fecal wet weight (FWW) and X-rayed to count the radioopaque pellets. They were then homogenized as a composite of the 5-d period, and dry matter (FDM) and ash (FA) were determined by methods previously described (5, 14). Mean values for the 5-d periods were used in all calculations.

The ash was analyzed by means of an Instrumentation Laboratory Model IL200 Inductively Coupled Plasma (ICP) Emission Spectrometer (Allied Analytical Systems, Andover, MA). The ICP power level was 1.4 kw and the sample introduction rate was 1 ml/min. The following wave lengths were used: Ca, 422.67; Mg, 279.08; Fe,

238.20; Zn, 213.86; and Cu, 324.75 nm.

Fecal steroids were extracted from the fecal homogenate with toluene after digestion with acetic acid. After hydrolysis of conjugated bile acids by treatment with cholyglycine hydrolase, neutral steroids were extracted with petroleum ether at pH 10 and acidic steroids with ethyl acetate and ethyl ether after reducing the pH to 1.0. Neutral steroids were quantitated by gas-liquid chromatography as trimethyl silyl ethers by using 3% OV-17 on 100/120 mesh Supelcoport (Supelco, Bellefonte, PA) and by using 5 α -cholestane as an internal standard. Bile acids were quantitated similarly as methyl acetates using 3% SP-2100 on 100/200 mesh Supelcoport and 23-nordeoxycholic acid as internal standard (15-17).

Statistical analyses. The data were analyzed by using a 3-way analysis of variance, unbalanced design, with period, subject, and treatment as the factors (18). The Statistical Package for the Social Sciences (SPSS) (19) program was used to perform all statistical analyses.

RESULTS

Period effect and compliance. Differences between period 1 and period 2 were found to be not statistically significant for the variables analyzed ($P > 0.05$); thus the means for each treatment were calculated

TABLE 2
Incomplete block design for treatment distribution¹

Subject no.	Period 1 treatment	Period 2 treatment
1, 13, 25	A	B
2, 14, 26	C	D
3, 15, 27	A	C
4, 16, 28	B	D
5, 17, 29	A	D
6, 18, 30	B	C
7, 19, 31	B	A
8, 20, 32	D	C
9, 21, 33	C	A
10, 22, 34	D	B
11, 23, 35	D	A
12, 24, 36	C	B

¹A, no hard red wheat bran (HRWB); B, 13.2 g/d HRWB; C, 39.6 g/d HRWB; D, 66.0 g/d HRWB.

from data from both periods. One subject dropped out for reasons unrelated to the study, thus groups A and B have 17 instead of 18 subjects.

Daily review of dietary records showed that the typical food intake did not significantly change for any of the subjects during the study. All subjects consumed treatments as required under the supervision of a study monitor.

Fecal weight and ash. Each increase in dietary HRWB induced a statistically significant increase in FWW, FDM and FA (table 3). Percent fecal dry matter (FDM%) did not change significantly between groups A, B and C, but there was a statistically significant ($P < 0.05$) drop at the highest level of HRWB (66.0 g/d).

Fecal minerals. Total daily excretion of fecal minerals increased for calcium, magnesium, iron, zinc and copper with increased HRWB (table 4). When the amount of minerals supplied by the wheat bran was subtracted from the total daily fecal mineral excretion, only calcium showed a significant ($P < 0.05$) increased excretion per day, i.e., there was no increased loss of magnesium, iron, zinc or copper caused by added HRWB.

Transit time. Mouth-to-anus TT, as shown in table 3, decreased significantly ($P < 0.05$) after addition of 13.2 g HRWB (group A to B) but did not significantly change after the further addition of 26.4 g HRWB (group B to C). TT decreased significantly again for the highest level of HRWB cor-

responding to a further addition of 26.4 g HRWB (group C to D) ($P < 0.05$).

Fecal steroids. Total daily fecal steroid excretion was not significantly altered by HRWB intake (table 5). Concentration of fecal total steroids was reduced 21, 50 and 64% by addition of HRWB to the diet, the two high levels causing a statistically significant reduction ($P < 0.05$). Similar changes were seen in neutral steroid excretion (table 5). Bile acid concentration in feces was not changed at the lowest level of HRWB intake but was reduced 13 and 36% by the higher levels of intake. Daily bile acid excretion was increased by intake of HRWB (38, 65 and 60%), the two higher levels of intake causing significant increases in bile acid excretion.

In examining the changes in daily bile acid excretion (table 6), the increase can be seen to result primarily from an increase in daily excretion of chenodeoxycholic, a primary bile acid, which increased 44, 99 and 110% with the three levels of HRWB. These increases were statistically significant at the two highest intakes of HRWB ($P < 0.05$). Daily excretion of the other bile acids also tended to increase but none of the changes were statistically significant or quantitatively as important as the changes in chenodeoxycholic acid. In addition the ratio of primary (cholic and chenodeoxycholic acids) to secondary bile acid (lithocholic and deoxycholic acids) increased from 0.62 in the group with no added HRWB to 1.14 for 66 g/d of HRWB (table 6).

TABLE 3

Effect of three levels of hard red wheat bran (HRWB) and of a diet free from wheat bran on gastrointestinal transit time, fecal wet and dry weight, moisture and ash^{1,2}

Measure	Group and HRWB intake, g/d			
	A 0	B 13.2	C 39.6	D 66.0
n	17	17	18	18
Transit time, d	3.18 ± 0.30 ^a	2.43 ± 0.24 ^b	2.44 ± 0.36 ^b	2.05 ± 0.21 ^c
Fecal wet wt, g/d	73.4 ± 6.3 ^a	94.7 ± 10.0 ^b	139.5 ± 11.6 ^c	212.0 ± 16.6 ^d
Fecal dry wt, g/d	20.0 ± 1.5 ^a	25.1 ± 2.2 ^b	34.0 ± 2.6 ^c	48.8 ± 3.2 ^d
Fecal dry wt, %	27.5 ± 0.74 ^a	27.49 ± 1.16 ^a	25.07 ± 0.93 ^{ab}	23.86 ± 0.95 ^b
Fecal ash, g/d	2.66 ± 0.18 ^a	3.36 ± 0.31 ^b	4.91 ± 0.39 ^c	6.93 ± 0.49 ^d

¹Values are 5-d means ± SEM. ²Values without a common superscript are significantly different ($P < 0.05$).

TABLE 4

Effect of three levels of hard red wheat bran (HRWB) and of a diet free from wheat bran on fecal mineral before and after subtracting the amount supplied by HRWB¹

Measure	Group and HRWB intake, g/d			
	A 0	B 13.2	C 39.6	D 66.0
n	17	17	18	18
Total daily excretion, mg/d				
Calcium	470 ± 51 ^a	530 ± 65 ^a	658 ± 69 ^b	755 ± 67 ^b
Magnesium	135 ± 10.4 ^a	196 ± 18.2 ^a	341 ± 31.0 ^b	547 ± 34.0 ^c
Iron	12.3 ± 1.7 ^a	16.9 ± 4.1 ^a	18.3 ± 1.6 ^b	22.7 ± 2.1 ^b
Zinc	4.55 ± 0.44 ^a	5.56 ± 0.59 ^a	8.17 ± 0.89 ^b	11.52 ± 1.02 ^c
Copper	1.05 ± 0.08 ^a	1.13 ± 0.09 ^a	1.62 ± 0.16 ^b	2.32 ± 0.32 ^c
Excretion minus intake from HRWB, mg/d				
Calcium	470 ± 51 ^a	522 ± 65 ^a	635 ± 69 ^b	717 ± 67 ^c
Magnesium	135 ± 10.4 ^a	117 ± 18.2 ^a	103 ± 31.0 ^a	151 ± 34.0 ^a
Iron	12.3 ± 1.7 ^a	14.7 ± 4.1 ^a	11.8 ± 1.6 ^a	11.8 ± 2.1 ^a
Zinc	4.55 ± 0.44 ^a	4.04 ± 0.59 ^a	3.62 ± 0.89 ^a	3.93 ± 1.02 ^a
Copper	1.05 ± 0.08 ^a	0.93 ± 0.09 ^b	1.01 ± 0.16 ^b	1.30 ± 0.32 ^{ab}

¹Values are 5-d means ± SEM. ²Values without a common superscript are significantly different ($P < 0.05$).

DISCUSSION

The addition of 29.0 g (table 3) of cereal fiber caused an increase in FWW from 73.4 ± 6.3 in HRWB-free group to 212.0 ± 16.5 g/d, corresponding to about 4 g increase in FWW per gram of DF. This is similar to the increase of approximately 4 g FWW per g DF found by Cummings et al. (20) for wheat bran from crispbread and is greater than the effect of purified cellulose of 2.7 g/d (14). The FWW of the low DF group (group A) is similar to those of puri-

fied, fiber-free diets, i.e., the 70–90 g/d range (21) showing that wheat and similar fiber are very effective in increasing fecal weight among common foods (1).

The fact that the decrease in FDM% was significant only at the highest level of HRWB (table 3) might well be related to the faster mean TTs of 2.05 d.

There is a significant decrease in TT between the HRWB-free diet and group B (lowest level of HRWB intake); no significant change appears between groups B and C ($P > 0.05$) indicating that intakes of

TABLE 5

Effect of three levels of hard red wheat bran (HRWB) and of a diet free from wheat bran on fecal steroid concentration and excretion^{1,2}

Group	HRWB g/d	n	Neutral steroids		Acidic steroids		Total (neutral + acidic) steroids	
			Concn mg/g	Daily excretion mg/d	Concn mg/g	Daily excretion mg/d	concn mg/g	Daily excretion mg/d
A	0	17	30.8 ± 3.9 ^b	641 ± 86 ^a	2.82 ± 0.34 ^b	57.8 ± 7.9 ^a	33.6 ± 4.1 ^b	700 ± 89 ^a
B	13.2	17	23.6 ± 3.4 ^b	568 ± 72 ^a	2.87 ± 0.33 ^b	79.8 ± 13.3 ^{ab}	26.4 ± 3.5 ^b	648 ± 75 ^a
C	39.6	18	14.4 ± 1.9 ^a	550 ± 93 ^a	2.49 ± 0.29 ^{ab}	95.6 ± 13.9 ^b	16.8 ± 2.0 ^a	645 ± 101 ^a
D	66.0	18	10.5 ± 2.0 ^a	492 ± 65 ^a	1.80 ± 0.20 ^a	92.5 ± 9.2 ^b	12.3 ± 2.1 ^a	585 ± 64 ^a

¹Values are 5-d means ± SEM. ²Values without a common superscript are significantly different ($P < 0.05$).

TABLE 6

Effect of three levels of hard red wheat bran (HRWB) and of a diet free from wheat bran on concentration and excretion of fecal bile acids^{1,2}

Bile acid measure ³	Group and HRWB intake, g/d			
	A 0	B 13.2	C 39.6	D 66.0
n	17	17	18	18
Lithocholic, mg/g	0.42 ± 0.06 ^a	0.33 ± 0.06 ^a	0.32 ± 0.05 ^a	0.21 ± 0.07 ^a
mg/d	7.9 ± 1.3 ^a	8.7 ± 1.4 ^a	11.2 ± 1.8 ^a	9.3 ± 2.3 ^a
Deoxycholic, mg/g	1.31 ± 0.17 ^b	1.18 ± 0.18 ^b	0.98 ± 0.13 ^{ab}	0.60 ± 0.10 ^a
mg/d	25.7 ± 4.4 ^a	30.0 ± 19.7 ^a	37.0 ± 7.1 ^a	30.0 ± 3.4 ^a
Chenodeoxycholic, mg/g	1.02 ± 0.13 ^a	1.01 ± 0.18 ^a	1.12 ± 0.14 ^a	0.78 ± 0.09 ^a
mg/d	19.9 ± 2.8 ^a	28.6 ± 7.4 ^{ab}	39.7 ± 25.3 ^b	41.7 ± 5.2 ^b
Cholic, mg/g	0.05 ± 0.02 ^a	0.09 ± 0.04 ^a	0.05 ± 0.01 ^a	0.05 ± 0.01 ^a
mg/d	0.8 ± 0.3 ^a	3.4 ± 2.2 ^a	1.9 ± 0.5 ^a	3.1 ± 0.6 ^a
Others, ⁴ mg/g	0.18 ± 0.05 ^a	0.26 ± 0.09 ^a	0.16 ± 0.03 ^a	0.15 ± 0.03 ^a
mg/d	3.5 ± 0.9 ^a	9.1 ± 3.9 ^a	5.7 ± 1.3 ^a	8.5 ± 2.1 ^a

¹Values are 5-d means ± SEM. ²Values without a common superscript are significantly different ($P < 0.05$). ³Where milligrams/gram is concentration in feces and milligrams/day is total daily excretion.

⁴Hyodeoxycholic, ursodeoxycholic, α -, β - and ω -muricholic acids detected but not in all samples.

HRWB in the range of 13–40 g/d induce similar TT, and a further significant change takes place when the HRWB is increased to 66 g/d ($P < 0.05$).

When the minerals naturally present in HRWB were subtracted from the fecal minerals (table 4), it appeared that the increase in HRWB did not cause any increased loss of magnesium, iron, zinc and copper. The only increased loss was for calcium, which increased from 470 to 717 mg/d, corresponding to 247 mg/d. This may suggest that calcium intake should be increased when diets high in wheat bran are eaten, especially for high risk groups such as pregnant women and the elderly, although adaptation seems to occur in the long term in people consuming high fiber diets (22). The increased fecal calcium corresponds to approximately 200 ml of milk. It is important to remember that the HRWB had been fermented with yeast during bread making and that these results may be applicable only to wheat bran as part of yeast-fermented breads, in which the phytate content has been modified by the fermentation process (6, 7). As this was not designed as a mineral balance study, the results for mineral excretion must be interpreted with caution.

The main change in fecal steroid excretion in response to an increase in HRWB is a decrease in concentration of both neutral and acidic steroids. This change has also been observed in response to other dietary fiber sources and has clinical significance in the relationship between fecal steroid concentrations, especially bile acids, and colon cancer susceptibility (23, 24).

The increase in daily excretion of bile acids indicates some effect of HRWB on bile acid metabolism. This increase (table 6) was due to an increase in primary bile acid excretion, while secondary bile acid excretion did not change. Other sources of dietary fiber have been suggested to cause such increases by adsorption of bile acids (8), but the lack of increase in fecal bile acid concentration tends to deny this mechanism. Since the change is relatively small in comparison to total fecal steroid excretion, it is unlikely to cause any direct change in cholesterol balance. However, if the increase in chenodeoxycholic acid, a primary bile acid, reflects changes in the bile acid pool sizes, reductions in cholesterol synthesis and absorption (25) could influence cholesterol balance. Indeed Munoz et al. (26) have previously reported reduced serum cholesterol in response to HRWB in humans. More

information is needed before these relationships can be understood.

In conclusion, the results of this study show that: 1) transit times do not decrease linearly as dietary fiber increases; 2) fecal moisture increases significantly only at a fairly high level of wheat fiber intake; 3) fecal mineral excretion increases only for calcium, not for zinc, iron, copper or magnesium when baker's yeast-fermented hard wheat bran is used, although these results must be interpreted in the light of an uncontrolled mineral intake; 4) there is an alteration in the type of acidic steroid excretion in response to wheat fiber increases, while total fecal steroid excretion does not change. The fecal weight and transit time data from this dose-response study should be useful in the determination of a desirable intake of wheat-type fiber for adults, when these data are added to the pool of data previously published on this subject (1).

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