β -Glucan-Enriched Fractions from Laboratory-Scale Dry Milling and Sieving of Barley and Oats^{1,2}

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ABSTRACT

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Dry milling and sieving of barley and oats produced fractions that were enriched in β -glucan [(1 \rightarrow 3), (1 \rightarrow 4)- β -D-glucan]. Hulled and hull-less varieties of barley and defatted rolled oats and oat bran, containing less than 10% moisture, were ground and sieved with a 325-mesh screen (45- μ m openings). Coarse material retained by the screen was reground and resieved. The resulting coarse fraction, with a weight yield of 18-30%, contained 16-27% total β -glucan. Further sieving of this β -glucan-rich

fraction with a 100-mesh screen (147- μ m openings) yielded fractions representing 1.7-16.5% of the kernel weight and containing up to 28% total β -glucan. The water-soluble β -glucan was about 55% of the total β -glucan for all test materials except Wanubet barley, in which soluble β -glucan ranged from 60 to 70% of the total β -glucan. Dry milling and sieving techniques can be used to prepare barley and oat fractions with β -glucan concentrations 2.4-4.9 times those of the original grain.

Cereal grains contain β -glucan, which influences digestion (Gohl 1977, Gohl et al 1978, White et al 1981) and cholesterol level in blood and liver tissue (Fadel et al 1987, Klopfenstein and Hoseney 1987, Welch et al 1988, Newman et al 1989, Wood et al 1989a). Diets that include grains with a high β -glucan content may be healthful since lowering of cholesterol by the β -glucan may decrease the risk of heart attack.

Barley and oats contain a higher level of β -glucan than other cereals and are thus suitable starting materials for preparing β -glucan concentrates. β -Glucan concentrates (80% β -glucan) can be prepared from barley by extraction with water followed by precipitation with alcohol (B. E. Knuckles, *unpublished results*), as described in the analytical method of Åman and Graham (1987). Wood et al (1989b) prepared an oat fraction containing 78% β -glucan by extracting a defatted air-classified oat bran with sodium carbonate.

However, β -glucan fractions prepared by extraction and precipitation are too costly for commercial use in foods. Dry milling would be a more economical way to obtain grain fractions enriched in β -glucan. Grinding and sieving (Bach Knudsen and Eggum 1984) and grinding and air classification (Danielson et al 1989) have been used to prepare barley fractions containing 6-23% β -glucan. This paper describes a simple laboratory-scale drymilling process that gives good yields of β -glucan-rich fractions from barley and oats.

MATERIALS AND METHODS

Grains Tested

We compared two hulled (Steptoe and Klages) and two hulless (CI 4362 and Wanubet) varieties of barley with respect to yield and β -glucan enrichment of fractions obtained by dry milling and sieving. Steptoe and Klages were chosen because they are available commercially and represent feed and malting varieties. CI 4362 and Wanubet were chosen because they do not require a dehulling step and potentially contain more β -glucan. Unfortunately, Wanubet, which was bred to contain extra-high levels (9+%) of β -glucan, contained less β -glucan than expected. Rolled oats and oat bran (full-fat and defatted) from a local store were also included in the study because oats are known to have high levels of β -glucan.

The hull was removed from Steptoe and Klages barley kernels

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with an abrasive disk mill (Satake Engineering Co. Ltd., Tokyo, Japan) fitted with a screen with openings measuring 1×25 mm. Grain (100 g) was milled for 1 min. A small sample was also dehulled by hand for comparison.

Oat samples (250 g) were defatted by soaking (22–25°C, 20 hr) and rinsing in *n*-hexane (1 L each). Solids were separated from hexane by centrifugation (10,000 \times g, 10 min). Hexane was removed from the solids by passing a stream of air across the material in an evaporation dish.

Milling and Sieving

We tested a variety of grinding mills and standard sieves (20-cm-diam., W. S. Tyler Co., Cleveland, OH) on a rotary-taping shaker (Ro-Tap, W. S. Tyler Co.) during the development of the process for preparing β -glucan-enriched fractions. Mills that were tested included a ball (jar) mill (Norton, Akron, OH), a pin mill (Alpine model 160Z, Hosokawa Micron International, Summit, NJ), an abrasive stone disk mill (Meadows Mill Co., North Wilkesboro, NC), a roller mill (C. W. Brabender Instruments, Inc., South Hackensack, NJ), and an abrasive mill (0.25-mm screen, Udy Corporation, Fort Collins, CO). Sieves (20-cm-diam.) were 100-, 200-, and 325-mesh, with openings of 147, 75, and 45 μ m, respectively.

Figure 1 is a diagram of the process that was used to prepare β -glucan-rich fractions from barley and oat materials. Unless noted otherwise, sieving was for 1 hr. Processing of each test material was replicated on different days.

Analytical Methods

Moisture was measured as weight lost upon heating at 85°C for 20 hr. β -Glucan was determined by enzymatic procedures based on those of McCleary and Glennie-Holmes (1985) and McCleary and Nurthen (1986). The method involves measuring glucose with glucose oxidase-peroxidase reagent after digestion with lichenase (EC 3.2.1.6) and β -glucosidase (EC 3.2.1.21). Enzymes, glucose reagent, and procedures were supplied in a kit (K-133, Biocon, Lexington, KY).

Soluble and insoluble β -glucan were separated in a manner similar to that reported by Aman and Graham (1987). Material (100 mg), in tared centrifuge tubes, was suspended in water (5.0 ml), held at 38°C for 2 hr, and centrifuged (2,300 \times g, 15 min). The pellet was washed twice with 5.0 ml of water. The washed pellet was suspended in phosphate buffer (50 mM, pH 4.0), and insoluble β -glucan was determined as for total β -glucan. Soluble β -glucan in the combined water extract was precipitated with ethanol (final concentration 70%), recovered by centrifugation (2,300 \times g, 5 min), and analyzed as for total β -glucan.

Starch was determined by a modification of AACC Method 76-11 (AACC 1983). The ground sample (0.1 g) was weighed into a tared 50-ml centrifuge tube. Water (25.0 ml) was added with stirring, and pH was adjusted to 5-7. The suspension was boiled for 3 min, then autoclaved (121°C) for 1 hr. When the

sample was removed from the autoclave, 20 ml of acetate buffer (0.5M, pH 4.8) was added to the mixture for a total solution weight of 45 ± 1 g. Enzyme solution (5.0 ml) (150 IU of amyloglucosidase [EC 3.2.1.3; Sigma Chemical Co., St. Louis, MO] and 20 IU of α -amylase [EC 3.2.1.1; Sigma] in 30 mM CaCl₂) was added to the mixture, which was then held at 50°C overnight. Reducing sugars were measured by adding 50 μ L of supernatant $(2,300 \times g, 15 \text{ min})$ to 3.0 ml of cyanide reagent (KCN [0.2 g] and K₃Fe(CN)₆ [0.19 g] in 200 ml of 2% Na₂CO₃), incubating for 5 min (100°C), and measuring absorbance at 420 nm.

Glucose in β -glucan and starch analysis was calculated from regression equations for three levels of glucose standard versus absorbance. The correlation coefficients (r^2) for glucose versus absorbance in the assays for β -glucan and starch were 0.99+ and -0.99+, respectively. Glucose, after subtraction of blank glucose values, was converted to β -glucan or starch by multiplying by 0.9. The blank for β -glucan was as described in the assay kit. For the starch blank, sample was treated as in the assay except no enzymes were added.

Duplicate fractionations were performed on each test material. Each analysis for each fraction was performed in duplicate (different days). Data are reported on a dry weight basis (db). Statistical measurements included standard error of mean (SEM), variance, and Tukey's HSD multiple pairwise comparisons. These analyses were performed with Systat software (Systat Inc, Evanston, IL).

RESULTS AND DISCUSSION

Milling and Sieving

Steptoe and Klages barleys have hulls that were removed by hand or in an abrasive disk mill. The hull removed by hand from Steptoe and Klages barleys was 13.4 ± 0.9 and $11.0 \pm 0.1\%$, respectively, of whole kernel weight. Hull fractions removed by disk milling these barleys for 1 min were 14.5 ± 0.4 and $11.5 \pm 0.2\%$, respectively, of the original weight. (Visual inspection showed very little hull remaining on kernels.) These data suggest that Steptoe has a thicker hull than Klages and that hulls can be satisfactorily removed by mechanical means.

Before selecting a grinder to use in this study, we ground dehulled Steptoe barley in various mills and compared the weight and the β -glucan and starch contents of each fraction that passed through a 325-mesh screen (45- μ m openings). With the ball mill, roller mill, pin mill, abrasive stone disk mill, and abrasive Udy mill, 79, 35, 49, 30, and 70%, respectively, of the dehulled barley weight passed through the screen. All these fractions contained less than 1.8% β -glucan and 70-80% starch. We chose the Udy mill because it allowed both rapid grinding of small samples and the separation of large amounts of material with a low β -glucan content.

We chose the 325-mesh screen for use in the process because

it allowed passage of starch granules, the major component separable from β -glucan. Starch granules are less than 40 μ m in diameter (Hoseney 1986). When the ground barley was sieved through a stacked series of screens (100-, 200-, and 325-mesh), 20 hr was required for complete sieving. Using the 325-mesh screen alone reduced sieving time to only 1 hr. Apparently, larger particles are needed to prevent clogging of the 325-mesh screen.

The sequence of steps we followed for β -glucan enrichment and the letter designations of the resultant fractions are shown in Figure 1. The yields of the fractions produced with this fractionation scheme are given in Table I. Recovery of materials at

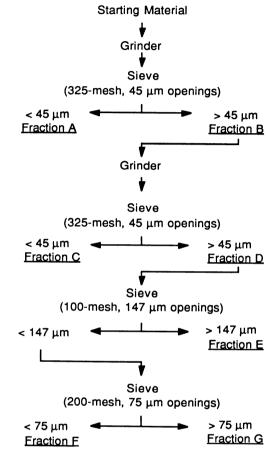


Fig. 1. Grinding and sieving of barley and oat materials. Starting material was dehulled Steptoe and Klages barleys, hull-less barleys CI 4362 and Wanubet, and defatted rolled oats and oat bran. Starting material (40-50 g) was ground in a Udy sample mill (0.25-mm screen) and sieved for 1 hr on a Ro-Tap sieve shaker.

TABLE I
Weight (%) of Fractions from Laboratory-Scale Milling and Sieving of Barley and Oats^a

Milling Fraction	Particle Size (µm)	Barley				Oats	
		Steptoe ^b	Klages ^b	CI 4362°	Wanubet	Rolled	Brane
Starting							
Material		100.0	100.0	100.0	100.0	100.0	100.0
Α	<45	72.1 ± 0.1	73.1 ± 0.4	50.6 ± 1.1	71.1 ± 0.8	63.2 ± 1.4	72.2 ± 0.3
В	>45	27.6 ± 1.5	26.8 ± 0.4	48.9 ± 0.6	29.1 ± 0.8	36.9 ± 1.8	28.5 ± 0.1
C	<45	6.7 ± 0.3	7.7 ± 0.4	19.0 ± 0.8	4.2 ± 0.2	18.6 ± 1.0	5.7 ± 0.2
D	>45	20.7 ± 0.2	18.7 ± 0.4	30.1 ± 0.2	25.0 ± 1.2	18.3 ± 1.0	23.0 ± 0.6
E	>147	2.7 ± 0.2	2.1 ± 0.1	2.2 ± 0.2	4.5 ± 0.2	5.2 ± 0.5	1.7 ± 0.1
F	>75	12.3 ± 0.5	10.7 ± 0.1	16.5 ± 0.3	11.7 ± 0.5	7.3 ± 0.4	11.4 ± 0.3
G	<75	5.9 ± 0.1	6.0 ± 0.1	11.7 ± 0.4	8.8 ± 0.4	5.9 ± 0.3	9.7 ± 0.5

^a Dry weight basis. Data are means (± SEM) of two determinations. Moisture content of ground Steptoe, Klages, CI 4362, Wanubet, rolled oats, and oat bran was 7.8, 7.2, 4.7, 5.5, 5.5, and 4.1%, respectively.

^b Hull removed from Steptoe and Klages kernels was 14.5 and 11.5%, respectively.

^c Hull-less varieties.

^d Defatted with *n*-hexane.

^e Defatted with *n*-hexane. Sieved for 2 hr because screen tended to clog.

each step in processing was excellent (100 \pm 0.5%). Fraction A from the various materials, with the exception of CI 4362, contained more than 63% of the starting weight (in the case of Steptoe and Klages, dehulled weight). A much smaller amount (4.2–19.0%) of the starting weight was in fraction C. Yield of β -glucan-rich fraction D was 18–30% of the starting weight. These yields tend to be higher than the approximately 16% yield of a high- β -glucan fraction obtained from barley by air classification (A. D. Danielson, personal communication). Vose and Youngs (1978) reported similar yields (16%) of a coarse barley fraction from air classification, but β -glucan concentration was not determined.

When full-fat rolled oats were ground and sieved with the 325-mesh screen, only 0.16% of the weight passed through the screen; thus, no enrichment of β -glucan was obtained. Rolled oats and oat bran fractionated after fat was removed yielded 18.3 and 23%, respectively, of the starting weight as fraction D. These yields for fraction D from defatted oat materials were similar to the 21% yield of a β -glucan-rich fraction obtained by Wood et al (1989b), who fractionated oats using a procedure that included ethanol deactivation and ethanol sieving (150- μ m bolting cloth). Wood et al (1989b) also demonstrated that 57% of pinmilled full-fat oat groats would pass a 355- μ m bolting cloth.

Milling reduced 69-82% of the test materials to particles of less than 45 μ m (fractions A and C). The remaining fraction D was subjected to a limited particle size distribution using 100-and 200-mesh screens, resulting in fractions E, F, and G (Fig. 1). Results show that 6-28% of fraction D, or 2-5% of the original weight, was composed of particles larger than 147 μ m, and 40-60% of the weight in fraction D was composed of particles larger than 75 μ m but smaller than 147 μ m (fraction G).

Moisture content could affect fractionation of materials: high moisture could result in stickiness, and very low moisture could allow development of a static charge. Static charge, as indicated by material behavior, was observed during sieving. This effect was most evident with defatted oat bran and may have contributed to the longer sieving time required for oat bran.

β-Glucan Content

Removal of the hull, which contains less than 0.6% of total β -glucan, enriched the total β -glucan concentration of Steptoe and Klages barleys by 18 and 45%, respectively (Table II). Total β -glucan concentrations in fractions D-G produced from these dehulled barleys were 3.0-4.3 times higher than those of the unfractionated materials. Total β -glucan concentrations in similar fractions from hull-less barleys were only 2.3-3.0 times higher, mainly because the hull-less barleys contained more β -glucan to begin with.

In general, 47-55% of the total β -glucan in fractions of Steptoe, Klages, and CI 4362 was soluble. Where solubility of β -glucan exceeded this range, errors may have been caused by incomplete sedimentation of insoluble glucans during analysis. Solubility of β -glucan was 10-15% higher in Wanubet barley than in other barleys.

Fraction E had the highest β -glucan concentrations of fractions from all barleys. The 18.6–22.5% total β -glucan for these fractions exceeds the 6% reported for other sieved barley fractions (Fadel et al 1987) and approaches the 23% reported for an air-classified fraction of barley (Danielson et al 1989).

Soluble β -glucan was 50, 40, 51, and 67% of total β -glucan in unfractionated Steptoe, Klages, CI 4362, and Wanubet barley, respectively. These solubilities are within the 38-69% range re-

TABLE II β -Glucan Content (%) of Barley and Oat Fractions^a

Fraction and		В	Oats			
β-Glucan	Steptoe ^b	Klages ^c	CI 4362 ^d	Wanubet ^d	Rolled	Bran ^e
Starting material						
Total	$5.3 c \pm 0.1$	$5.1 c \pm 0.1$	$6.8 \text{ b} \pm 0.1$	$7.2 \text{ b} \pm 0.1$	$4.7 e \pm 0.1$	$9.6 a \pm 0.4$
Insoluble	$2.4 c \pm 0.1$	$2.3 c \pm 0.1$	$3.2 \text{ b} \pm 0.1$	$2.5 c \pm 0.1$	$2.2 d \pm 0.1$	$3.9 a \pm 0.3$
Soluble	$2.6 d \pm 0.2$	$2.4 d \pm 0.1$	$3.5 c \pm 0.1$	$4.8 b \pm 0.1$	$2.3 d \pm 0.1$	$5.7 a \pm 0.3$
Fraction A						
Total	$1.7 c \pm 0.1$	$1.6 c \pm 0.2$	$1.2 c \pm 0.1$	$2.0 \text{ b} \pm 0.1$	$0.6 d \pm 0.1$	$4.2 a \pm 0.2$
Insoluble	$0.7 \text{ b} \pm 0.1$	$0.7 \text{ b} \pm 0.1$	$0.5 \text{ b} \pm 0.1$	$0.6 \text{ b} \pm 0.1$	$0.3 \text{ b} \pm 0.1$	$1.4 a \pm 0.1$
Soluble	$0.9 c \pm 0.1$	$0.8 c \pm 0.1$	$0.6 c \pm 0.1$	$1.4 \text{ b} \pm 0.1$	$0.1 c \pm 0.1$	$1.7 a \pm 0.1$
Fraction B						
Total	$14.9 c \pm 0.8$	$14.2 c \pm 0.5$	$11.4 d \pm 0.6$	$19.5 \text{ b} \pm 0.3$	$12.5 d \pm 0.3$	$22.6 \text{ a} \pm 1.8$
Insoluble	$6.6 \text{ b} \pm 0.1$	$6.1 \text{ bc} \pm 0.3$	$5.4 d \pm 0.3$	$6.6 \text{ b} \pm 0.1$	$5.7 \text{ cd} \pm 0.3$	$11.6 \text{ a} \pm 1.5$
Soluble	$7.6 \text{ b} \pm 0.1$	$6.4 c \pm 0.3$	$6.0 c \pm 0.3$	$12.8 \text{ a} \pm 0.1$	$6.5 \text{ bc} \pm 0.3$	$11.7 \text{ a} \pm 0.9$
Fraction C						
Total	$4.9 c \pm 0.2$	$5.7 c \pm 0.4$	$6.0 c \pm 0.5$	$10.3 \text{ b} \pm 0.1$	$4.5 d \pm 0.2$	$12.0 \text{ a} \pm 0.3$
Insoluble	$2.0 \text{ b} \pm 0.1$	$2.3 \text{ b} \pm 0.2$	$2.7 \text{ b} \pm 0.2$	$2.5 b \pm 0.1$	$1.6 c \pm 0.2$	$3.3 a \pm 0.3$
Soluble	$2.5 c \pm 0.1$	$2.8 c \pm 0.3$	$3.0 c \pm 0.3$	$7.1 \text{ b} \pm 0.3$	$0.5 d \pm 0.1$	$7.6 \text{ a} \pm 0.1$
Fraction D						
Total	$17.6 \text{ d} \pm 0.6$	$17.6 d \pm 0.6$	$16.0 \text{ d} \pm 0.3$	$19.9 c \pm 0.3$	$21.2 \text{ b} \pm 0.6$	$27.2 \text{ a} \pm 1.7$
Insoluble	$7.6 c \pm 0.1$	$7.9~\mathrm{c}\pm0.2$	$7.6 c \pm 0.1$	$6.6 d \pm 0.2$	$9.4 \text{ b} \pm 0.4$	$12.9 \text{ a} \pm 1.4$
Soluble	$9.2 d \pm 0.3$	$8.6 d \pm 0.1$	$8.5~{ m d}\pm0.2$	$14.8 \text{ a} \pm 0.8$	$11.0 c \pm 0.4$	$13.8 \text{ b} \pm 0.7$
Fraction E						
Total	$22.5 \text{ cd} \pm 1.2$	$22.0 \text{ cd} \pm 1.1$	$18.6 \text{ d} \pm 0.2$	$21.3 c \pm 0.4$	$23.2 b \pm 0.6$	$28.1 \text{ a} \pm 0.5$
Insoluble	$8.4~\mathrm{d}\pm0.3$	$9.4 c \pm 0.1$	$10.1 c \pm 0.1$	$8.2 d \pm 0.1$	$12.3 \text{ b} \pm 0.5$	$13.6 \text{ a} \pm 0.5$
Soluble	$13.0 \text{ b} \pm 0.9$	$13.1 \text{ b} \pm 1.2$	$8.7 e \pm 0.3$	$15.6 \text{ a} \pm 0.1$	$10.4 \ d \pm 0.3$	$12.8 c \pm 0.7$
Fraction F						
Total	$17.4 c \pm 0.5$	$17.1 c \pm 0.9$	$16.0 c \pm 0.4$	$18.6 c \pm 0.1$	$23.6 \text{ b} \pm 0.1$	$26.4 \text{ a} \pm 2.5$
Insoluble	$8.0 c \pm 0.1$	$7.9 c \pm 0.1$	$8.0 c \pm 0.2$	$7.1 d \pm 0.1$	$10.2 \text{ b} \pm 0.3$	$15.0 \text{ a} \pm 2.0$
Soluble	$9.1 c \pm 0.3$	$8.3 c \pm 0.3$	$8.4 c \pm 0.1$	$15.1 \text{ a} \pm 0.1$	$13.0 \text{ b} \pm 0.4$	$14.0 \text{ ab} \pm 2.1$
Fraction G						
Total	$16.8 \text{ bc} \pm 0.5$	$15.3 c \pm 0.5$	$15.8 c \pm 0.3$	$18.1 \text{ b} \pm 0.1$	$17.2 \text{ bc} \pm 0.3$	$25.3 \text{ a} \pm 1.7$
Insoluble	$7.2 \text{ b} \pm 0.2$	$7.3 \text{ b} \pm 0.5$	$7.2 \text{ b} \pm 0.1$	$5.5 c \pm 0.1$	$6.2 \text{ bc} \pm 0.3$	$9.1 \text{ a} \pm 0.7$
Soluble	$7.6 \text{ e} \pm 0.3$	$7.7 e \pm 0.3$	$8.7 d \pm 0.1$	$13.1 \text{ b} \pm 0.1$	$10.4 c \pm 0.3$	$16.1 \text{ a} \pm 0.7$

^a Values are means \pm SEM (duplicate samples, duplicate analyses on each) reported on dry weight basis. Within each row, values followed by the same letter are not significantly different (P < 0.05).

^b Dehulled. With the hull, total β -glucans were 4.5% and soluble β -glucans were 2.25%.

^c Dehulled. With the hull, total β -glucans were 3.5% and soluble β -glucans were 1.4%.

d Hull-less varieties.

^e Defatted with *n*-hexane.

TABLE III
Starch Content (%) of Barley and Oat Fractions^a

		Bar	Oats			
Fraction	Steptoe ^b	Klages	CI 4362 ^d	Wanubet ^d	Rolled	Brane
Starting material	$70.3 \text{ a} \pm 0.7$	$70.7 \text{ a} \pm 0.5$	61.9 b ± 0.4	64.3 b ± 0.4	$72.2 \text{ a} \pm 1.8$	$56.9 c \pm 0.9$
A	$79.2~{ m ab}\pm 1.1$	$77.9 \text{ b} \pm 1.6$	$72.5 c \pm 0.9$	$70.9 c \pm 1.6$	$80.1 \text{ a} \pm 0.7$	$61.0 d \pm 0.9$
В	$52.4 b \pm 0.7$	$53.1 \text{ b} \pm 0.9$	$51.9 \text{ b} \pm 1.8$	$53.7 \text{ b} \pm 0.4$	$61.6 \text{ a} \pm 0.3$	$45.0 \text{ c} \pm 0.9$
C	$67.3 \text{ b} \pm 0.3$	$66.1 \text{ b} \pm 1.1$	$54.9 c \pm 2.0$	$56.7 c \pm 1.6$	$73.9 \text{ a} \pm 0.7$	$51.4 c \pm 0.7$
D	$49.4 \text{ ab} \pm 0.6$	$49.9 \text{ ab} \pm 1.0$	$47.2 \text{ b} \pm 0.7$	$50.7 \text{ a} \pm 0.8$	$52.2 \text{ a} \pm 1.6$	$45.2 \text{ b} \pm 1.0$
E	$62.6 \text{ b} \pm 1.8$	$75.0 \text{ a} \pm 2.0$	$69.5 a \pm 1.8$	$64.0 \text{ b} \pm 3.4$	$50.7 c \pm 2.2$	$45.1 c \pm 1.8$
F	$48.9 \text{ b} \pm 0.5$	$48.2 \text{ b} \pm 1.0$	$45.9 b \pm 1.3$	$55.4 a \pm 0.2$	$47.0 \text{ b} \pm 0.6$	$41.9 c \pm 0.9$
G	$47.1 c \pm 0.6$	$47.4 c \pm 0.6$	$42.0 d \pm 1.0$	$59.7 a \pm 1.3$	$56.7 \text{ b} \pm 0.9$	$44.9 \text{ cd} \pm 0.9$

^a Values are means \pm SEM (duplicate samples, duplicate analyses on each) reported on a dry weight basis. Within each row, values followed by the same letter are not significantly different (P < 0.05).

ported for a number of other barley varieties (Åman and Graham 1987). Solubility of β -glucan in dehulled Steptoe and Klages was 49 and 47%, respectively, and was similar in rolled oats (49% soluble). Solubility of β -glucan in oat bran (59%) was higher than in rolled oats but lower than in Wanubet barley. The higher level of soluble β -glucan in oat bran probably results from prior processing, whereas the higher level in Wanubet barley probably results from breeding for high β -glucan content.

Fractionation of oat materials produced fractions containing up to 28% total β -glucan (Table II). In most of these fractions, 50-60% of the β -glucan was soluble. However, soluble β -glucan was higher than this in fractions C and G, possibly because of errors in analysis due to incomplete sedimentation of waterinsoluble β -glucan. (In general, separation of solubles and insolubles was more difficult with oat materials than with barley.)

Total β -glucan concentration in oat fractions D-G exceeded the 11-16.6% in fractions prepared by Wood et al (1989b). Solubility (44-64%) of β -glucan in β -glucan-enriched fractions from oats was lower than the range (65-90%) reported by Åman and Graham (1987).

For each material except rolled oats, fraction E had the highest β -glucan concentration. This fraction was also composed of the largest particles (larger than 147 μ m). This suggests that β -glucan is associated with materials that are more resistant to size reduction than starch. β -Glucan has been demonstrated to be in endosperm cell walls and inner walls of aleurone cells in oats and barley (Fulcher and Wood 1983, Wood et al 1983).

Except for Wanubet barley, with about 90% recovery of insoluble β -glucan at the second sieving, and oat bran, with about 90% recovery of soluble β -glucan at all processing steps, calculated recoveries for total, insoluble, and soluble β -glucan at each fractionation step were better than 95%. Recovery of soluble β -glucan at the second sieving of Wanubet was 104%, and recoveries of insoluble β -glucan during fractionation of oat bran were 103–107%. Thus, the low recovery of insoluble β -glucan at the second sieving of Wanubet and of soluble β -glucan during fractionation of oat bran may be the result of difficulties in centrifugal separation of insoluble and soluble β -glucan. Although temperature, pH, and endogenous enzyme activity may greatly affect β -glucan extraction yields (Åman and Graham 1987), these factors did not appear to affect β -glucan recoveries in this study.

Starch Content

Starch concentration in whole barley ranged from 61.9% in CI 4362 to 66.1% in Klages (Table III). Removal of the hull from Steptoe and Klages barley resulted in a starting material for fractionation that had a higher starch content than hull-less barleys and defatted oat bran. Rolled oats had a much higher starch content than defatted oat bran because of prior separation of bran and starchy endosperm.

Fraction A had the highest starch content of all fractions from all starting materials. This was expected since starch granules, being less than 45 μ m in diameter, passed through the 325-mesh sieve. Fraction A contained 81.2, 80.5, 59.3, 78.4, 70.1, and 77.4% of total starch present in unfractionated Steptoe, Klages, CI 4362, Wanubet, rolled oats, and oat bran, respectively. Smaller amounts of total starch (6.4, 7.1, 16.9, 3.7, 19.0, and 5.1%, respectively) were recovered in fraction C. The $100 \pm 2\%$ recoveries of starch for all materials at all fractionation steps indicate that starch was not lost as a result of processing or endogenous enzymes.

 β -Glucan-rich fraction D contained a relatively high concentration of starch (45–52%) (Table III), suggesting that further grinding and sieving could further increase β -glucan concentration. The fact that fraction E (particles larger than 147 μ m) from barleys contained a higher level of starch than fractions F and G (with smaller particles) is evidence that additional starch could be removed during barley fractionation. However, further processing to remove additional starch must be balanced against the fact that more β -glucan would also be removed. (Note that β -glucan concentrations in C fractions are more than twice those in A fractions [Table II]. This difference suggests that further grinding and sieving [325-mesh] would result in fractions with particles less than 45 μ m in diameter having higher concentrations of β -glucan.)

The high starch content and low β -glucan content of fraction A give this fraction potential for enhanced utilization of oats and barley in particular. The high starch content would make the material attractive as a starch source or an adjunct for beer production. Low glucan content lessens the deleterious effects associated with normal to high levels of glucan (Hesselman et al 1982, Bamforth 1985, McCleary and Glennie-Holmes 1985, Newman and Newman 1985, Hesselman and Åman 1986, Klopfenstein and Hoseney 1987).

CONCLUSIONS

These small-scale studies demonstrate the possibility of producing barley and oat fractions that are substantially enriched in β -glucan. The simple dry-milling process produces fractions high in β -glucan (up to 28%) and in yields up to 30% of the initial weight. Oat materials were more difficult to fractionate than barley. Fractionation of oats was successful only after removal of fat. Most of the material being separated from β -glucan appeared to be starch. The ability to prepare products with differing β -glucan and starch content could increase the economic value of barley and oats by opening the possibility of tailoring fractions for specific markets.

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b Hulled variety, hull removed. With the hull, Steptoe contained 64.4% starch.

^c Hulled variety, hull removed. With the hull, Klages contained 66.1% starch.

d Hull-less varieties.

^e Defatted with n-hexane.

LITERATURE CITED

- ÅMAN, P., and GRAHAM, H. 1987. Analysis of total and insoluble mixed-linked (1→3),(1→4)-β-D-glucans in barley and oats. J. Agric. Food Chem. 35:704.
- AMERICAN ASSOCIATION OF CEREAL CHEMISTS. 1983. Approved Methods of the AACC. Method 76-11, approved October 1976, reviewed October 1982. The Association: St. Paul, MN.
- BACH KNUDSEN, K. E., and EGGUM, B. O. 1984. The nutritive value of botanically defined mill fractions of barley. 3. The protein and energy value of pericarp testa, germ, aleuron, and endosperm rich decortication of the variety Bomi. Z. Tierphysiol. Tierernaehr. Futtermittelkd. 51:130.
- BAMFORTH, C. W. 1985. Biochemical approaches to beer quality. J. Inst. Brew. 91:154.
- DANIELSON, A. D., McGUIRE, C. F., NEWMAN, R. K., NEWMAN, C. W., and SCHWARZ, P. B. 1989. Dietary fiber content of air classified fractions of hull-less waxy barley. Barley Newsl. 33:147.
- FADEL, J. G., NEWMAN, R. K., NEWMAN, C. W., and BARNES, A. E. 1987. Hypocholesterolemic effects of beta-glucans in different barley diets fed to broiler chicks. Nutr. Rep. Int. 35:1049.
- FULCHER, R. G., and WOOD, P. J. 1983. Identification of cereal carbohydrates by fluorescence microscopy. Pages 111-147 in: New Frontiers in Food Microstructure. D. B. Bechtel, ed. Am. Assoc. Cereal Chem.: St. Paul, MN.
- GOHL, B. 1977. Influence of water treatment of barley on digestion process in rats. Z. Tierphysiol. Tierernaehr. Futtermittelkd. 39:57.
- GOHL, B., ALDEN, S., ELWINGER, K., and THOMKE, S. 1978. Influence of β-glucanase on productive value of barley and on moisture content of poultry excreta. Br. Poult. Sci. 19:41.
- HESSELMAN, K., and AMAN, P. 1986. The effect of β -glucanase on the utilization of starch and nitrogen by broiler chickens fed on barley of low-viscosity or high-viscosity. Anim. Feed Sci. Technol. 15:83.
- HESSELMAN, K., ELWINGER, K., and THOMKE, S. 1982. Influence of increasing levels of β-glucanase on the productive value of barley diets for broiler chickens. Anim. Feed Sci. Technol. 7:351.

- HOSENEY, R. C. 1986. Cereal starch. Pages 33-68 in: Principles of Cereal Science and Technology. Am. Assoc. Cereal Chem.: St. Paul, MN.
- KLOPFENSTEIN, C. F., and HOSENEY, R. C. 1987. Cholesterollowering effect of beta-glucan-enriched bread. Nutr. Rep. Int. 36:1091.
- McCLEARY, B. V., and GLENNIE-HOLMES, M. 1985. Enzymic quantification of $(1\rightarrow 3)$, $(1\rightarrow 4)-\beta$ -D-glucan in barley and malt. J. Inst. Brew. 91:285.
- McCLEARY, B. V., and NURTHEN, E. J. 1986. Measurement of (1-3), $(1-4)-\beta$ -D-glucan in malt, wort, and beer. J. Inst. Brew. 92:168.
- NEWMAN, R. K., and NEWMAN, C. W. 1985. Beta-glucanase effect on the performance of broiler chicks fed covered and hull-less barley isotypes having normal and waxy starch. Proc. Am. Soc. Anim. Sci. West. Sect. 36:239.
- NEWMAN, R. K., NEWMAN, C. W., and GRAHAM, H. 1989. The hypocholesterolemic function of barley β-glucans. Cereal Foods World 34:883.
- VOSE, J. R., and YOUNGS, C. G. 1978. Fractionation of barley and malted barley flours by air classification. Cereal Chem. 55:280.
- WELCH, R. W., PETERSON, D. M., and SCHAMKA, B. 1988. Hypocholesterolemic and gastrointestinal effects of oat bran fractions in chicks. Nutr. Rep. Int. 38:551.
- WHITE, W. B., BIRD, H. R., SUNDE, M. L., PRENTICE, N., BURGER, W. C., and MARLETT, J. A. 1981. The viscosity interaction of barley beta-glucan with *Trichoderma viride* cellulase in the chick intestine. Poult. Sci. 60:1043.
- WOOD, P. J., FULCHER, R. G., and STONE, B. A. 1983. Studies on the specificity of interaction of cereal cell wall components with Congo Red and Calcofluor. Specific detection and histochemistry of (1→3), (1→4)-β-D-glucan. J. Cereal Sci. 1:95.
- WOOD, P. J., ANDERSON, J. W., BRAATEN, J. T., CAVE, N. A., SCOTT, F. W., and VACHON, C. 1989a. Physiological effects of β-D-glucan rich fractions from oats. Cereal Foods World 34:878.
- WOOD, P. J., WEISZ, J., FEDEC, P., and BURROWS, V. D. 1989b. Large-scale preparation and properties of oat fractions enriched in (1-3)(1-4)-β-D-glucan. Cereal Chem. 66:97.

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