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A Comparison between Detergent and Nondetergent Analyses of Dietary Fiber in Human Foodstuffs, Using High-Performance Liquid Chromatography To Measure Neutral Sugar Composition

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Dietary fiber analysis of wheat bran, peas, apples, and a food composite, using the modified neutral detergent fiber (NDF) method and a nondetergent method, has been examined. In the latter method, starch hydrolysis was enhanced by using ultrasonics for gelatinization and a heat-stable α-amylase. Neutral sugar compositions, determined by high-performance liquid chromatography, of the insoluble fractions were similar, although, with the exception of wheat bran, neutral detergent extracted significantly more arabinans than did water. NDF contents of the foodstuffs were marginally lower than the corresponding nondetergent fiber contents, due to lower concentrations of uronic acids and lignin in the NDF residues. Water-soluble fractions recovered by the nondetergent method represented only a small portion of the dry matter of the foodstuffs studied. However, for apples and food composite the fraction occupied about 25% of the total dietary fiber content.

Increasing evidence (Spiller and Kay, 1980) that dietary fiber (DF) has an important physiological role in human nutrition has resulted in a considerable increase in the number of published methods for measuring DF in human foodstuffs. Recently, many of the principal approaches were evaluated in an international collaborative study (James and Theander, 1981). The range of results obtained in this study show that uniform and accurate measurement of DF is intrinsically difficult and is complicated further by the wide variety of approaches currently being used (Asp and Johansson, 1981; Furda, 1981; Robertson and Van Soest, 1981; Southgate, 1981; Theander and Aman, 1981).

Direct analysis of the carbohydrate components in DF is necessary in order to conduct meaningful physiological studies and to compare the DF constituents of different foods. Colorimetric (Southgate, 1981) and gas-liquid chromatographic (Englyst, 1981; Theander and Aman, 1981) methods are commonly used to measure carbohydrates in DF. However, recent technological developments in carbohydrate analysis by high-performance liquid chromatography (HPLC) (McGinnis and Fang, 1980) have made it feasible to quantitate the major carbohydrate components in DF from human foodstuffs (Slavin and Marlett, 1983) by using this comparatively simple and rapid technique.

The modified neutral detergent fiber (NDF) method (American Association of Cereal Chemists, 1977; Robertson and Van Soest, 1981) has been used for food fiber analysis, although the method has been critized because it is gravimetric and only provides empirical information. This laboratory recently reported that measurement of the neutral sugars in acid hydrolysates of NDF residues by HPLC overcomes this disadvantage. The NDF method, however, does not recover the water-soluble fiber fraction that has been associated with physiologically significant functions (Anderson, 1980; Jenkins, 1980; Kay and Truswell, 1980). In contrast, the nondetergent procedure of

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Table I. Component Distribution in the Residues Isolated by the Nondetergent and Detergent Methods and the Recoveries from the Complete Residue Analysis[a]

sample	water-insoluble residues: starch, mg	protein,[b] mg	dietary[c] fiber, mg	recovery, %	water-soluble residues: starch, mg	protein,[b] mg	dietary[c] fiber, mg	recovery, %
wheat bran								
nondetergent	trace	354 ± 10	1128 ± 21	93.7 ± 3.6	8 ± 1	48 ± 13	51 ± 1	64.2 ± 3.7
neutral detergent	trace	36 ± 0	677 ± 4	94.7 ± 0.2				
peas								
nondetergent	41 ± 2	526 ± 38	592 ± 23	100.2 ± 3.4	13 ± 2	97 ± 11	90 ± 3	90.9 ± 0.9
neutral detergent	11 ± 1	trace	156 ± 4	97.1 ± 1.1				
apples								
nondetergent	trace	13 ± 1	358 ± 6	95.3 ± 1.4	trace	16 ± 9	107 ± 1[d]	93.5 ± 2.4
neutral detergent	trace	trace	141 ± 5	102.0 ± 5.0				
food composite								
nondetergent	7 ± 0	182 ± 27	177 ± 23	106.6 ± 4.9	32 ± 4	36 ± 1	57 ± 7	89.6 ± 6.6
neutral detergent	4 ± 0	6 ± 1	126 ± 10	109.3 ± 3.9				

[a] Mean ± SD, $n = 2$. [b] $N \times 6.25$. [c] Dietary fiber = original dry matter (mg) × dietary fiber content (%), calculated from the overall analytical procedure (Tables III–V). [d] This value includes an acid-insoluble precipitate representing 11.2% of the water-soluble residue isolated.

Theander and Aman (1979a, 1981), which is based on direct carbohydrate analysis, isolates both water-insoluble and water-soluble DF fractions. The conflicting data and views of various authors (James and Theander, 1981) and our own interests in fiber with respect to its physiological role in man prompted a comparison of these two diverse but relatively straightforward procedures. In particular, the yield and composition of the NDF fractions of four representative foods have been compared to those of the nondetergent-insoluble fractions. The composition and quantitative contribution of the water-soluble fraction to the total DF were also determined.

MATERIALS AND METHODS

Sample Preparation. Wheat bran (AACC-certified food-grade wheat bran, St. Paul, MN) was ground to 30 mesh in a Thomas-Wiley mill. Canned peas (Del Monte), fresh apples (Washington State Red Delicious) with the cores removed, and a food composite, previously described (Slavin and Marlett, 1980), were each blended with distilled water and lyophilized. Dry weights of prepared samples were determined by AOAC procedures of the Association of Official Analytical Chemists (1975).

Isolation of Water-Insoluble Fiber Fractions. Water-insoluble and water-soluble fractions were obtained by slightly modifying the procedure of Theander and Aman (1979a, 1981). Extractive-free samples were ultrasonicated in water (50 mL) at 85 °C for 30 min prior to incubation with an excess (0.2 mL) of the α-amylase, Termamyl 120 L (activity 120 KNU/g, batch AAP 0003, Novo Laboratories Inc., Wilton, CT). During the course of this work, we were informed that the Theander Laboratory had independently incorporated ultrasonication in their extraction procedure (Theander and Westerlund, 1982). Wheat bran and apples were separated into their respective fractions by Büchner vacuum filtration through filter paper (Whatman No. 54). No. 54 paper was used since it has a relatively fast filtering rate; however, the paper allowed small particulate matter (<23-μm particle size) to pass through. These fine particles were removed by centrifugation, 12000g, 5 min, and combined with the water-insoluble residue. A check for β-glycosidase activity was made by incubating xylan (Larch Wood, Aldrich) and α-cellulose (Sigma Chemical Co., St. Louis, MO) with the Termamyl enzyme under similar conditions and measuring reducing sugar concentrations (McFeeters, 1980). Substrates with added enzyme at 0-min incubation time were used as controls. The amounts of reducing sugars released by the enzyme were very small, 0.3 and 1.6% (by weight) of the α-cellulose and xylan, respectively. This amount of activity was not considered a significant source of error in the dietary fiber calculations.

Peas and food composite were fractionated by centrifugation, 12000g, 5 min, immediately after boiling. Particulate-free supernatants were aspirated off and the pellets washed with water (3 × 50 mL, 100 °C). An additional similar experiment was conducted using peas, but in this case centrifugation and washing were at 25 °C. Residues were then transferred to a Büchner funnel with ethanol. All insoluble residues were washed (Theander and Aman, 1981), vacuum dried (<40 °C, 5 h, P_2O_5), and weighed. Total filtrates and/or combined supernatant liquors were dialyzed (Spectra/Por 4 tubing, M_r cutoff 12000–14000, Spectrum Medical Industries, Inc., Los Angeles, CA), 48 h, 4 °C, against 4 × 4 L of deionized, glass-distilled water, lyophilized, vacuum dried and weighed. Neutral detergent fiber (NDF) residues were prepared by a modified NDF procedure used in this laboratory (Slavin and Marlett, 1983) that includes glass wool as a filtering aid (Brauer et al., 1981) and sodium sulfite. Starch was measured by the procedure of the American Association of Cereal Chemists (1976) and nitrogen by the Kjeldahl method (Fleck and Munro, 1965).

Determination of Neutral Carbohydrates by HPLC and Apparent Lignin. Nondetergent residues were hydrolyzed by modifying the procedure of Blake and Richards (1970). Insoluble and soluble residues (25–50 mg) were thoroughly mixed with 72% sulfuric acid (300 mg of residue/mL of acid) at 25 °C, kept in a desiccator with occasional stirring for 1 h. Water was added (28 mL/mL of acid) and secondary hydrolysis carried out under reflux for 3 h at 100 °C. Erythritol (5–10 mg) was then added as an internal standard. Where insoluble material was present, the mixture was centrifuged, 1000g, 10 min, and the hydrolysate aspirated into a second centrifuge tube for neutralization. The resultant pellet was washed free of acid with water, vacuum dried, weighed, and reported as apparent lignin (Theander and Aman, 1979a). Hydrolysates were neutralized at 60 °C with barium carbonate, centrifuged, 1000g, 10 min, and the supernatants filtered through a 0.2-μm filter. The NDF residues were hydrolyzed by a modification of the procedure of Saeman et al. (1954), as previously performed in this laboratory (Slavin and Marlett, 1983).

The sugar compositions of the hydrolysates were determined by HPLC (Slavin and Marlett, 1983). The chromatographic system has been essentially described (Slavin and Marlett, 1983). A microguard column (Aminex

HPX-85C, Bio-Rad, Richmond, CA) was in series with a carbohydrate analysis column (Aminex HPX-87P heavy metal, 300 mm × 7.8 mm, Bio-Rad) operated at 85 °C with filtered and degassed water, at a flow rate of 0.6 mL/min. The amounts of sugars present were computed by a reporting integrator after calibration data were obtained with vacuum-dried, standard sugars (Sigma). The component monosaccharides have been expressed as polysaccharides following correction for hydrolysis losses and residual starch if present. Recoveries from the modified hydrolytic procedure (Blake and Richards, 1970) were determined by subjecting authentic sugars to the total analytical procedure. Two mixtures each containing 20 mg of three different aldoses were analyzed in triplicate. The losses allowed for in calculation of the NDF component sugars have been published previously (Slavin and Marlett, 1983) and are as follows (%): glucose, 33.5; xylose, 38.1; galactose/rhamnose, 32.9; arabinose, 37.1; mannose, 25.6. Crystalline α-cellulose (25 mg) (Sigma) was subjected to hydrolysis to determine the accuracy of the overall hydrolytic procedure, except that 150 mg of polysaccharide/mL of acid was used for the primary acid hydrolysis. In addition, the apparent lignin residue from wheat bran was rehydrolyzed to check for complete hydrolysis; no sugars were detected in the hydrolysate.

Uronic Acid Analysis. Uronic acids were determined colorimetrically, by adapting the 3-hydroxydiphenyl (Pfaltz and Bauer, Inc., Stamford, CT) method of Blumenkrantz and Asboe-Hansen (1973) using D-galacturonic acid (Sigma) as the standard. Uronic acid yields were expressed as a polysaccharide. Water-soluble residues (5 mg) were dissolved in water and diluted prior to uronic acid analysis. Uronic acid containing polysaccharides were solubilized from non-detergent-insoluble residues with 72% sulfuric acid (Saeman et al., 1954; Slavin and Marlett, 1983). Residue (5 mg) was dispersed in 72% sulfuric acid (0.41 g) and allowed to stand for 3 h, 25 °C, in a desiccator with occasional stirring. The mixture was then diluted (to 6 mL) and gravity filtered through glass fiber paper [Whatman (GF/A)]. Acidic polysaccharides were solubilized from NDF residues on glass wool by using a similar 3-h acid treatment described earlier (Slavin and Marlett, 1983). Immediately following dilution, the mixture was vacuum filtered through glass fiber paper; any remaining residue and glass wool were washed with water. Recovery of D-glucuronic acid (5 mg) was determined after subjecting it to 72% sulfuric acid for 0 and 3 h at 25 °C.

RESULTS AND DISCUSSION

The use of ultrasonics for gelatinization and the heat-stable amylase improved the removal of starch from the nondetergent water-insoluble fiber residues. Following amylase treatment, the water-insoluble residue from peas contained 5.7% starch when static conditions were used for gelatinization and 3.5% starch with ultrasonics. Ultrasonics would be expected to have a positive effect on the enzymatic hydrolysis of starch, since the accompanying vigorous agitation facilitates the dissolution of polysaccharides (Pittet, 1965), and it has been used to obtain aqueous suspensions of cellulose microfibrils in structural studies on the polymer (Manley, 1964). Also, starch remaining in the water-insoluble residue from peas after Termamyl digestion was less, 3.5% of residue dry weight, than that remaining following amyloglucosidase (Sigma) treatment, 5.0%. The heat-stable Termamyl enzyme used for starch hydrolysis has an optimum temperature of ~85 °C, and compared to the widely used amyloglucosidase it is inexpensive and requires a shorter incubation time. Termamyl, an endoamylase specific for $\alpha 1 \rightarrow 4$ glucosidic

Table II. Effect of Fractionation Temperature on the Composition and Yield of the Fiber Fractions Isolated from Peas[a,b]

	% total neutral sugars						% original dry matter			
	cellobiose	glucose	xylose	galactose	arabinose	mannose	total neutral sugar content	uronic acids	apparent lignin	dietary fiber content
water-insoluble fraction										
25 °C	11.5 ± 0.6	63.2 ± 0.1	7.9 ± 0.3	ND[c]	17.5 ± 0.3	trace	12.5 ± 0.7	2.4 ± 0.0	3.5 ± 0.1	18.4 ± 0.7
>25 °C	10.7 ± 1.3	63.6 ± 3.0	8.5 ± 2.2	ND	17.3 ± 2.1	trace	12.6 ± 1.1	2.6 ± 0.1	4.0 ± 1.4	19.2 ± 1.0
water-soluble fraction										
25 °C		5.7 ± 0.0	12.2 ± 0.8	7.6 ± 0.9	76.6 ± 1.2	trace	1.4 ± 0.1	1.4 ± 0.0		2.8 ± 0.1
>25 °C		5.7 ± 0.7	12.0 ± 0.7	7.4 ± 0.6	75.0 ± 0.6	trace	1.4 ± 0.1	1.4 ± 0.0		2.8 ± 0.1

[a] Mean ± SD, n = 2. [b] Neutral sugars and uronic acids are expressed as polysacchrides. Glucose has been corrected for residual starch. [c] ND = not detected.

Table III. Composition and Yield of the Water-Insoluble Fiber Fractions[a,b]

	% total neutral sugars						% original dry matter			
sample	cellobiose	glucose	xylose	galactose	arabinose	mannose	total neutral sugar content	uronic acids	apparent lignin	dietary fiber content
wheat bran	4.1 ± 0.0	26.9 ± 0.7	45.8 ± 0.6	ND[c]	23.2 ± 0.1	trace	29.9 ± 0.6	1.9 ± 0.2	5.9 ± 0.1	37.7 ± 0.7
peas	11.5 ± 0.6	63.2 ± 0.1	7.9 ± 0.3	ND	17.5 ± 0.3	trace	12.5 ± 0.7	2.4 ± 0.0	3.5 ± 0.1	18.4 ± 0.7
apples	8.6 ± 1.0	42.9 ± 1.0	13.5 ± 0.8	6.1 ± 0.3	25.6 ± 0.8	3.5 ± 0.2	5.4 ± 0.0	2.4 ± 0.1	1.9 ± 0.1	9.7 ± 0.2
food composite	6.4 ± 0.5	52.2 ± 1.6	18.0 ± 3.5	11.2 ± 1.3	12.3 ± 0.1	trace	3.7 ± 0.7	0.7 ± 0.0	0.9 ± 0.0	5.3 ± 0.7

[a] Mean ± SD, n = 2. [b] Neutral sugars and uronic acids are expressed as polysaccharides. Glucose has been corrected for residual starch. [c] ND = not detected.

Table IV. Composition and Yield of the Water-Soluble Fiber Fractions[a,b]

	% total neutral sugars					% original dry matter		
sample	glucose	xylose	galactose	arabinose	mannose	total neutral sugar content	uronic acids	dietary fiber content
wheat bran	19.5 ± 3.0	53.5 ± 4.0	ND[c]	26.2 ± 0.4	trace	1.5 ± 0.1	0.2 ± 0.0	1.7 ± 0.0
peas	5.7 ± 0.0	12.2 ± 0.8	7.6 ± 0.9	76.6 ± 1.2	trace	1.4 ± 0.1	1.4 ± 0.0	2.8 ± 0.1
apples	22.7 ± 3.6	11.8 ± 3.5	16.9 ± 0.6	46.7 ± 4.8	trace	0.5 ± 0.0	2.0 ± 0.1	2.9 ± 0.1[d]
food composite	negligible	20.6 ± 0.6	36.0 ± 0.4	43.5 ± 0.3	ND	0.8 ± 0.1	0.9 ± 0.2	1.7 ± 0.2

[a] Mean ± SD, $n = 2$. [b] Neutral sugars and uronic acids are expressed as polysaccharides. Glucose has been corrected for residual starch. [c] ND = not detected. [d] This value includes an acid-insoluble precipitate representing 0.4% of the original dry matter or 11.2% of the water-soluble residue hydrolyzed.

linkages, removes starch by hydrolyzing it into soluble dextrins and oligosaccharides; thus, nondialyzable starch–glucose was still present in the water-soluble fractions of all food samples except apples (Table I). This residual starch represented only 2–3% of the original starch content of the foods analyzed. In studies of total dietary fiber measurement, it is recommended to determine residual starch in all isolated fractions; consequently, such starch would be detected and the appropriate correction then can be made. Using this approach, we elected to omit the additional amyloglucosidase step prior to the dialysis of the water-soluble fraction (Theander and Aman, 1981).

Peas and food composite were fractionated into water-insoluble and water-soluble components by centrifugation, as the consistency of the sample suspension precluded vacuum filtration at 25 °C. Hot filtration (90 °C) with a water-jacketed Büchner funnel was also unsuccessful. Since the effect of temperature on fractionation could possibly influence the relative yield and composition of the two fractions, the centrifugation and washing procedures were performed at 25 °C and at an elevated temperature; no significant differences in the fiber fractions of peas separated under these two conditions were observed (Table II).

Recoveries of the reference sugars from the total analytical procedure (including hydrolysis) applied to the nondetergent residues were as follows (mean % ± SD of three): glucose, 94.4 ± 1.3; xylose, 94.4 ± 3.3; galactose, 86.2 ± 5.0; rhamnose, 102.4 ± 1.0; arabinose, 94.9 ± 1.2; mannose, 94.6 ± 1.0. The accuracy of the procedure was measured by hydrolyzing crystalline α-cellulose and duplicate results showed an overall recovery of 98.5%: cellobiose, 13.4%; glucose, 73.8%; xylose, 4.8%; mannose, 2.1%; apparent lignin, 4.4%. The stability of uronic acids was tested by subjecting glucuronic acid to 72% sulfuric acid, 3 h, 25 °C; no significant loss was observed (99% recovery). Galacturonic acid has been reported to be stable under similar acid conditions (Selvendran et al., 1979).

The two analytical procedures were applied to four different types of foodstuffs. The neutral sugar analyses of the water-insoluble fractions obtained by the modified Theander and Aman method showed that wheat bran was composed of arabinoxylans and glucans, which together represented 80% of the fraction (Table III). The pea insoluble fiber consisted of glucans (50%), with other neutral polysaccharide constituents of arabinose and xylose also present; uronic acids accounted for 13% of the pea fraction. Apples and food composite insoluble fiber fractions were a complex mixture of polysaccharides composed mostly of glucans with pectins and associated substances such as arabinose and galactose evident. A significant portion (25%) of the apple fiber was uronic acids in addition to the large glucan concentration; mannose also was detected in measurable amounts in this fraction. The food composite contained a relatively small percentage of water-insoluble dietary fiber. However, the predicted complexity of the polysaccharide components is revealed by the relative amounts of xylose, galactose, and arabinose, in addition to the presence of uronic acids. Consistent with its morphology, wheat bran had the highest proportion of apparent lignin.

The water-soluble fraction of wheat bran consisted of arabinoxylans and glucans (Table IV). The remaining foods contained large amounts of pectic substances and associated polysaccharides as evidenced by the high uronic acid contents, 50–70% of the fractions, and appreciable amounts of arabinose and galactose. Xylose was also present in relatively smaller amounts in the water-soluble fiber fraction of these foods. Arabinose (77%) was a predominant component of the pea polysaccharides, while the food composite had a significantly larger concentration of galactans (36%) than did the other samples. It should be noted that during secondary hydrolysis of the apple water-soluble fraction, a precipitate formed that represented ~11% by weight of the residue hydrolyzed. This fraction is largely uronic acids (~70%), and formation of the precipitate in these acid conditions was probably a result of degradation and/or condensation reactions involving uronic acids (Haug and Larsen, 1962; Selvendran et al., 1979; Theander and Aman, 1981).

Comparisons between the neutral sugar analyses of NDF fractions (Table V) and the corresponding data obtained from the non-detergent-derived insoluble fractions (Table III) revealed that the sugars in corresponding fractions were present in essentially similar concentrations. However, with the exception of wheat bran, it appears that the neutral detergent solution extracted significantly more arabinans from the foods we analyzed than did water. The DF contents of the samples as determined by the NDF method were marginally lower than those values obtained by using the nondetergent approach. This observation is consistent with the ability of the detergent solution to solubilize portions of pectin and lignin (Robertson and Van Soest, 1981), and our results confirm this property. Lignin is attacked by sodium sulfite, a constituent of the detergent mixture included to remove protein. Alternately, the acid-insoluble residue, which was used as an estimate of Klason lignin by Theander and Aman (1979a), may contain small amounts of proteinaceous material. Except for wheat bran, which retained about the same amounts of uronic acids and lignin, the total neutral sugar content of the NDF fractions was slightly higher than for the non-detergent-insoluble fractions; however, in general, the yields are comparable. The main disadvantage of the NDF method is that it does not recover a water-soluble fiber fraction. Although some attempt has been made to separate this fraction from detergent (O'Neill and Selvendran, 1980;

Table V. Composition and Yield of the NDF Fractions[a,b]

sample	% total neutral sugars						% original dry matter			
	cellobiose	glucose	xylose	galactose	arabinose	mannose	total neutral sugar content	uronic acids	apparent lignin	dietary fiber content
wheat bran	3.9 ± 0.1	26.2 ± 0.3	44.2 ± 0.1	ND[c]	25.7 ± 0.0	trace	28.4 ± 0.6	1.5 ± 0.1	6.7 ± 0.4	36.6 ± 0.2
peas	10.7 ± 0.6	74.1 ± 0.1	5.5 ± 1.1	trace	7.5 ± 0.1	2.5 ± 0.8	14.9 ± 0.4	0.9 ± 0.0	negligible	15.8 ± 0.4
apples	7.0 ± 1.1	51.9 ± 1.6	12.9 ± 0.1	8.1 ± 0.0	14.9 ± 0.1	5.5 ± 0.4	6.9 ± 0.3	0.4 ± 0.0	negligible	7.3 ± 0.3
food composite	7.5 ± 0.5	58.3 ± 0.9	15.0 ± 0.6	6.5 ± 0.7	8.6 ± 1.1	4.3 ± 1.6	4.1 ± 0.4	0.2 ± 0.0	negligible	4.3 ± 0.4

[a] Mean ± SD, $n = 2$. [b] Neutral sugars and uronic acids are expressed as polysaccharides. Glucose has been corrected for residual starch. [c] ND = not detected.

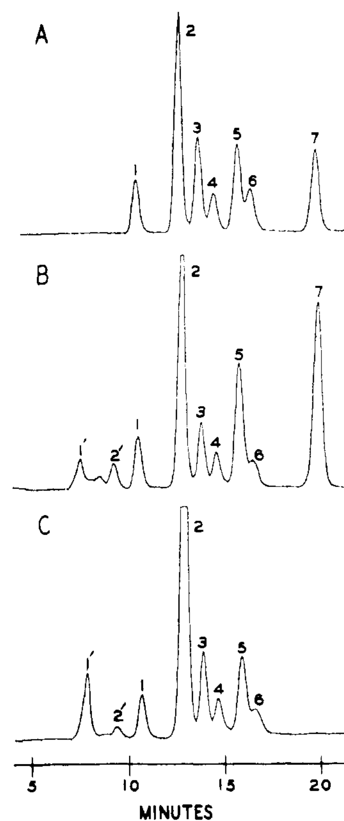

Figure 1. HPLC separation of constituent neutral sugars of dietary fiber on a Bio-Rad Aminex HPX-87P carbohydrate analysis column. Eluant, degassed water; 85 °C; 0.6 mL/min; refractive index indector. (A) Sugar standard mixture with peak designations: 1, cellobiose (0.05%); 2, glucose (0.20%); 3, xylose (0.10%); 4, galactose (0.05%); 5, arabinose (0.10%); 6, mannose (0.05%); 7, internal standard, erythritol (0.10%); attenuation 4X. (B) Apple, non-detergent-insoluble fiber; attenuation 4X. (C) Apple NDF; attenuation 8X.

Robertson and Van Soest, 1981), a reliable and reproducible isolation procedure needs to be developed.

The component sugars of the DF fractions were satisfactorily measured by HPLC (Figure 1). Peak 1′ is presumed to represent an acid degradation product of the neutral sugars since it was observed in the chromatograms of all authentic sugars subjected to the hydrolytic procedure. Peak 2′ was tentatively identified as cellotriose from its retention time relative to cellobiose, but in the absence of a reference compound it could not be unequivocally identified and quantitated. The Aminex HPX-87P carbohydrate analysis column we used could partly resolve galactose (retention time 14.54 min) from rhamnose (retention time 14.73 min); however, this partial resolution was not sufficient to detect the small amounts of rhamnose that may have been present in any of the foodstuffs analyzed in this study. Rhamnose has been detected in small amounts by using gas–liquid chromatography, in both fractions of peas and apples (Theander and Aman, 1979a). Although gas–liquid chromatography is more sensitive than HPLC with refractive index detection (McGinnis and Fang, 1980) for measuring carbohydrates, the speed and convenience of HPLC techniques far outweigh this disadvantage. Sugars present in small amounts may be measurable by concentrating hydrolysates prior to HPLC analysis.

The distribution of components in the isolated residues reflects the relative merits of the two approaches for determining DF in human foodstuffs (Table I). The ability of the NDF method to remove protein is clearly illustrated;

however, since this method is most widely used as a gravimetric technique, the presence of residual protein would overestimate the fiber content. The Theander and Aman approach does not require the quantitative removal of protein, since it involves direct carbohydrate determinations. Peas residues have the highest residual starch value, which is consistent with the routine finding in this laboratory that starch is difficult to remove from legumes (Marlett and Lee, 1980; Chesters and Marlett, 1982). Removal of the starch remaining in the soluble residues could possibly be obtained by more extensive dialysis. We are unable to account for the relatively low recovery of the wheat bran water-soluble fraction. Since the water-soluble fraction of human foodstuffs contributes significantly to the nutritional role of dietary fiber, this fraction will require further study. Nonetheless, the recoveries obtained from the total analytical procedures indicate that, overall, the starting material can be satisfactorily accounted for (Table I).

The complex chemical nature of total DF renders it difficult to define a distinct boundary between water-extractable constituents and the insoluble material (Theander and Aman, 1979b). Despite the heterogeneity of the fiber fractions recovered from the four foods in this study and the significant differences between the two methods, both the Theander and Aman and NDF methods produce similar yields of insoluble DF from human foodstuffs, and the neutral sugar compositions of the fractions were comparable, as well. The former procedure has the distinct advantage in that it recovers the water-soluble fiber fraction, although when Termamyl amylase is used, the fiber content of the soluble fraction must be corrected for any starch-derived oligosaccharides. Further, the Theander and Aman method of fiber analysis allows numerous analyses to be performed on a common fiber sample. In contrast, all of the NDF residue must be used for a single analysis. However, the NDF method is particularly useful for measuring the insoluble DF in fecal material, since it is specific for plant cell walls. The detergent dissolves microbially derived polymers (Robertson and Van Soest, 1981), leaving insoluble fiber that survives passage through the gut.

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