

# Fructan Content of Rye and Rye Products

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## ABSTRACT

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The fructan content of Finnish rye grains (13 samples, seven cultivars, harvested in 1998–2000) varied at 4.6–6.6 g/100 g (db). Commercial whole grain rye flour and rye flakes had fructan content of 4 g/100 g, light refined rye flour had fructan content of 3 g/100 g, and rye bran had fructan content of 7 g/100 g. Fructan content as high as 23 g/100 g was detected in the water-extractable concentrate of rye bran. Finnish soft rye

bread and rye crisp bread contained 2–3 g of fructan/100 g. According to the suggested new definition of dietary fiber, fructans are also classified as dietary fiber. This means that the dietary fiber content of some cereal foods such as rye products may be increased by as much as 20% due to the presence of fructans in the grain.

Fructans are  $\beta$ -D-linked polymers of fructose. Approximately 15% of higher plants store fructans (Ernst and Feldheim 2000). Good sources include Jerusalem artichoke (17–20 g/100 g), chicory (15–20 g/100 g), garlic (10–16 g/100 g), and onion (1–8 g/100 g, wb) (Van Loo et al 1995). Jerusalem artichoke and chicory are used as a source for commercial extraction of inulin. Fructans are present also in cereals, but literature on their occurrence in cereals is scarce.

Depending on the type of the linkage between fructosyl units, fructans can be divided into inulins, levans, and mixed levans (Vijn and Smeekens 1999). Inulin consists of linear (2-1)-linked  $\beta$ -D-fructosyl units and levan consists of linear (2-6)-linked  $\beta$ -D-fructosyl units. Mixed levan is composed of both (2-1)- and (2-6)-linked  $\beta$ -D-fructosyl units. The chain is mostly terminated by a glucose unit. Inulin, an extract of chicory root, as well as its enzymatic hydrolysis products, fructo-oligosaccharides, are commercially available food ingredients. In recent years, the prebiotic and bifidogenic properties, as well as the physiological responses, have been the subject of much scientific interest. Both inulin and fructo-oligosaccharides have improved gut function and selectively stimulated the growth of bifidobacteria (Bouhnik et al 1999; Brighenti et al 1999; Kruse et al 1999).

The fructan content of cereals is related to the content of water-soluble pentosans, and the amounts of both decrease in the order: rye, wheat, barley, oat, maize (MacLeod and Preece 1954). Rye, with a fructan content of 1.7–3.9%, appears to be the best cereal source of fructan (MacLeod and Preece 1954; Henry and Saini 1989). Content of fructan in wheat, barley, and oat is 0.9, 0.8, and 0.1 g/100 g, respectively (MacLeod and Preece 1954; Henry and Saini 1989). Later, higher amounts were found in wheat (1.3–2.5 g/100 g) (B. Fretzdorff, T. Kuhlmann, and T. Betsche, unpublished).

The fructans of rye have a higher degree of polymerization (DP) than those of wheat and barley. In rye, 78% of fructans have DP > 5 (Henry and Saini 1989). Fructans of low DP (<6) made up a substantial part ( $\approx$ 50%) of the fructans in wheat (Dahlqvist and Nilsson 1984; Nilsson and Dahlqvist 1986). White and Secor (1953) extracted fructans from wheat and found two structurally different series of fructans. Later, Medcalf and Cheung (1971) determined the primary D-fructose linkage in the major fraction to be  $\beta$ -2,6, and in the other fractions it was  $\beta$ -1,2.

Wheat fructans are indigestible in the small intestine (Nilsson and Björck 1988; Nilsson et al 1988), so they act in a manner similar to traditional dietary fiber (DF) and provide substrates for colon microflora. According to the suggested new definition of DF, fructans are also classified as DF (Anonymous 2001a). In general, rye, and especially rye bran, is a good source of DF. In addition to

fructans, the main fermentable DF components are the cell wall polysaccharides, arabinoxylans, and  $\beta$ -glucans. Soluble arabinoxylan and  $\beta$ -glucan, as well as commercial inulin, are fermented quickly during in vitro fermentation, whereas water-insoluble arabinoxylan of rye is fermented slowly (Karppinen et al 2000, 2001). Many of the health effects of DF are believed to be related to the microbial fermentation of these polysaccharides in the large intestine.

Rye is mostly eaten as bread. The most typical rye bread in Finland is whole grain rye bread made using the sour dough method. Another popular type of rye bread is crisp bread, which can be produced with or without sour dough fermentation. Fructan content has been reported to decrease during baking of rye (Boskov Hansen et al 2002) and wheat breads (Escrivá and Martínez-Anaya 2000). The aim of this study was to examine the level of fructan in Finnish rye grain, in some rye products, and also in a water-extractable concentrate of rye bran.

## MATERIALS AND METHODS

### Rye Samples

Samples of different Finnish rye cultivars were donated by S. Hovinen, Boreal Plant Breeding Ltd, Jokioinen, Finland. The cultivars were grown at the same locations in Jokioinen in three years, and were harvested in 1998–2000. Rye grains were stored at controlled temperature (18°C). Commercially available rye products included whole-grain rye flour, light refined rye flour (from which the outer part of the kernel was removed), rye flakes, rye crisp bread, and soft rye bread. Commercial rye bran Sieppari was obtained from Melia Ltd (Raisio, Finland) for the pilot-scale extraction.

Extraction of rye fructans was made in pilot scale to get enough material for in vitro and in vivo fermentation studies. Extruded rye bran (160 kg) and water (1,500 L) were mixed in a pilot fermenter at 50°C. Xylanase (Pentopan Mono BG, Novo, 300 g) was added to increase the amount of soluble components. The suspension was stirred at 50°C for 2 hr and then cooled. The solid material was allowed to settle overnight under cooling. The supernatant ( $\approx$ 650 L) was first decanted and then centrifuged (Alfa-Laval, Sweden), evaporated using a falling film evaporator (MTK-PE-1,1,6, Finland), and lyophilized in a pilot freeze dryer (Atlas, Denmark).

### Fructan Analyses

Rye grains, bran, flakes, and rye crisp bread were milled before analysis to pass through 0.5-mm sieve. Soft rye bread was homogenized. Fructan was extracted with hot water (80°C) and the content was determined by a specific enzymatic kit (Megazyme, Ireland) (McCleary et al 2000). The standard deviation of the fructan content of the control flour in the fructan kit (Megazyme), containing 29.3% dahlia fructan and made in every determination, was 3.5% ( $n = 31$ ). Fructan content of rye bran was measured also according to Method 997.08 (AOAC 1995) with a slight modi-

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fication so that extracts (made with hot water) were filtered before enzyme treatments. Samples of rye grain of 1998 and 1999 were analyzed in the beginning of 2000, and samples of 2000 were analyzed in the beginning of 2001. The fructan content of one rye flour and one rye crisp bread was analyzed as six replicates; standard deviation of the method was 5%. The rest of the analyses were performed as duplicates.

The fructo-oligosaccharide composition was determined by HPLC (DX 500 system, Dionex). Rye grain was milled to pass through a 0.5-mm sieve. Samples were extracted with water at room temperature, and 1-kestose, 1,1-kestotetraose, and 1,1,1-kestopentaose were quantified by peak area using commercial standards (Megazyme). The column was CarboPac PA1 (Dionex). The eluent was 2.5 mM NaOH up to 23 min and after that, it was gradually raised to 100 mM over 17 min. The flow rate was 1 mL/min and the oven temperature was 30°C.

### Other Chemical Analyses

Total starch and  $\beta$ -glucan contents were determined by specific enzymatic kits (Megazyme). Dietary fiber was measured according to Asp et al (1983). The pentosan content was measured according to Douglas (1981). Free sugars were determined from the water-extractable rye bran concentrate by the same liquid chromatographic method as fructo-oligosaccharides with the standards of sucrose, maltose, arabinose, xylose, glucose, and fructose. The protein content was determined by the Kjeldahl method ( $6.25 \times N$ ). Fat was determined according to the Method 922.06 (AOAC 1995). Ash content was determined by incineration overnight at 550°C using a temperature-programmed muffle oven. Results are mean values from two replicate determinations.

## RESULTS

### Fructan Content of Rye and Rye Products

The fructan content of three different cultivars of Finnish rye (Akusti, Anna, and Bor 7068) of 1998 was 6.4–6.6 g/100 g (db), as analyzed by the enzymatic method (Table I). The same three cultivars harvested in 1999 and 2000 had fructan content of 4.6–5.1 g/100 g and 4.8–5.4 g/100 g, respectively. Four more cultivars from 1999 (Riihi, Picasso, Voima, and Esprit) had fructan content of 4.6–5.9 g/100 g.

The fructan content of commercial whole grain rye flour analyzed enzymatically was 4.4–4.5 g/100 g (Table II). The fructan content of rye flakes was a little lower, 3.7–4.2 g/100 g. Rye bran had the highest fructan content (6.6 g/100 g) and light refined rye flour had the lowest content (3.1 g/100 g). According to the AOAC Method, the fructan content of rye bran was 6.7 g/100 g. The fructan content was 2.1–2.8 g/100 g for five Finnish soft rye breads and 2.2–2.6 g/100 g for two rye crisp breads.

### Water-Extractable Concentrate of Rye Bran

Extrusion and xylanase treatment were used as an aid to solubilize cell wall structures and, in this way, to increase the recovery of soluble components. Extrusion and xylanase pretreatment increased the content of soluble arabinoxylans and arabinoxylo-oligosaccharides in the extract approximately fourfold. The fructan content was also increased by the pretreatments, but the effect was not as great as in arabinoxylans (results not shown). Using this method, 9.4 kg of water-extractable concentrate was obtained from 160 kg of rye bran.

The fructan content of the water-extractable concentrate was as high as 23 g/100 g. The total carbohydrate content of rye bran was 71 g/100 g, and that of the water-extractable concentrate was 79 g/100 g (Table III). Pentosan and  $\beta$ -glucan contents were 23 and 3.5 g/100 g in rye bran and 28 and 4.7 g/100 g in the water-extractable concentrate, respectively. Rye bran contained 37 g of dietary fiber/100 g, as analyzed by Asp et al (1983) and water-extractable concentrate contained only 11 g/100 g. The sucrose

content of the water-extractable concentrate was 5.8 g/100 g and traces of free monosaccharides were detected.

### Fructo-Oligosaccharide Content

The fructo-oligosaccharide content of rye grain (cultivar Anna) was 0.6 g of 1-kestose/100 g, 0.3 g of 1,1-kestotetraose/100 g, and 0.3 g of 1,1,1-kestopentaose/100 g. In the water-extractable con-

TABLE I  
Fructan Content of Finnish Rye Cultivars in 1998-2000 (g/100 g, db)<sup>a</sup>

Cultivar	1998	1999	2000
Akusti	6.4	4.6	4.9
Anna	6.5	4.8	5.4
Bor 7068	6.6	5.1	4.8

<sup>a</sup> Results are mean values from two replicate determinations.

TABLE II  
Fructan Content of Various Commercial Rye Products (g/100 g, db)  
Analyzed Enzymatically

Product <sup>a</sup>	Fructan (g/100 g)
Brans, flakes, flours	
Rye bran	6.6
Rye flakes, 1	4.2
Rye flakes, 2	3.7
Rye flour, 1	4.5
Rye flour, 2	4.4
Rye flour, light refined	3.1
Breads	
Rye crisp bread, 1	2.6
Rye crisp bread, 2	2.2
Soft rye bread, 1	2.5
Soft rye bread, 2	2.8
Soft rye bread, 3	2.3
Soft rye bread, 4	2.1
Soft rye bread, 5	2.6

<sup>a</sup> Soft rye bread 1 contained whole grain rye (36%) and wheat flour, bread 2 contained whole grain rye (59%), bread 3 contained whole grain rye (70%) and wheat flour, bread 4 contained whole grain rye (71%) and no added yeast, and bread 5 contained rye flour.

TABLE III  
Composition (g/100 g, db) of Rye Bran and Water-Extractable  
Concentrate of Rye Bran

	Rye Bran	Water-Extractable Concentrate of Rye Bran
Basic composition		
Protein	17.5	9.7
Fat	4.6	2.4
Ash	6.5	8.6
Carbohydrate (as a difference)	71.4	79.3
Carbohydrate composition		
Fructan, total	6.6	22.6
Fructo-oligosaccharides		
1-kestose	nd <sup>a</sup>	6.0
1,1-kestotetraose	nd	0.4
1,1,1-kestopentaose	nd	1.0
Pentosan	22.7	28.1
Soluble pentosan	1.7	nd
$\beta$ -Glucan	3.5	4.7
Starch	25.4	12.8
Free sugars		
Sucrose	nd	5.8
Maltose	nd	1.8
Arabinose + xylose + glucose + fructose	nd	1.3
Other carbohydrates (cellulose, etc.)	13.1	2.2
Dietary fiber <sup>b</sup>	37.0	11.3
Soluble dietary fiber	4.5	11.3
Insoluble dietary fiber	32.4	0

<sup>a</sup> Not determined.

<sup>b</sup> According to Asp et al (1983).

centrate of rye bran, fructo-oligosaccharides were concentrated and the content was 6.0 g of 1-kestose/100 g, 0.4 g of 1,1-kestotetraose/100 g, and 1.0 g of 1,1,1-kestopentaose/100 g (Table III).

## DISCUSSION

Finnish rye grains analyzed in this study contained 4.6–6.6 g of fructan/100 g. The samples from three different years had different fructan contents. The highest fructan contents were in the 1998 samples. The annual variation may be mainly explained by differences in the growth temperature. In plants generally, fructans may have a function in protecting plants against water deficit caused by drought or low temperatures (Vijn and Smeekens 1999). Samples of both 1998 and 1999 were analyzed in the beginning of 2000 in the same determination, so samples of 1998 had longer storing time than samples of 1999. It is unlikely, however, that storage would have induced such an increase in fructan content.

The results of the present study showed higher levels for the fructan content of rye grain than were recorded in some earlier studies. MacLeod and Preece (1954) recorded 3.9 g of fructan/100 g of whole grain rye. According to Henry and Saini (1989), whole grains of rye had fructan contents of 1.7 g/100 g, and according to Glitsø (1997), whole grains of rye contained 2.5 g of fructan/100 g. The fructan content of 25 rye samples (five cultivars, five locations, harvest 1998) was enzymatically analyzed (B. Fretzdorff, T. Kuhlmann, and T. Betsche, *unpublished*) and the results were 4.8–7.4 g/100 g, which is in close agreement with the results presented here. Both methods, enzymatically according to a commercial kit (Megazyme) and the AOAC method, gave similar results for rye bran.

The fructan content of rye bran (6.6 g/100 g) is slightly greater than that of whole grain, indicating that fructan is accumulated in the outer parts of rye kernel. This confirms previous findings by Glitsø (1997), who reported that the highest fructan content was in the aleurone layer of the kernel. B. Fretzdorff, T. Kuhlmann, and T. Betsche (*unpublished*) analyzed milling fractions from rye grain. The fructan content was 2.6–3.3 g/100 g in the flours, and the highest fructan contents were determined in the shorts (7.2 g/100 g).

When eating whole grain rye, which is traditional in Finland, the fructan in the outer parts of the kernel is also available for human nutrition. If the outer part of rye kernel is removed in the milling process, as in light refined rye flour, the fructan content is lower (Table II). Whole grain rye is the main component in Finnish breads, but the amount varies from bread to bread. Also, wheat flour is used in some rye breads. Rye breads in Finland are usually made using the sour dough method, with or without added yeast. Boskov Hansen et al (2002) found that during breadmaking, where whole grain rye flour was used without any wheat flour, the content of fructan decreased from 6.2 g/100 g in the whole meal to 3.4 g/100 g in bread crumb. The differences between the breads (2.1–2.8 g/100 g) analyzed in this study are thus a consequence of the differences in the amounts of rye flour used, in the fructan content of the flours, and in differences in the fermentation conditions. Rye flakes contained a lower amount of fructan than rye grain. This may be a consequence of the processing conditions.

The main sources of daily intake of cereal fructans are bread and porridge. One slice of soft rye bread (25 g) contains 0.3–0.4 g of fructan according to the results presented in this study, and one slice of rye crisp bread (12 g) contains 0.3 g of fructan. Whole grain rye flakes are used in porridge in Finland, and one portion of porridge contains 1.2 g of fructan.

According to the Food Balance Sheet for Food Commodities 1999 and 2000, the average rye intake in Finland in 1999 and 2000 was 43 g/day (Anonymous 2001b), which corresponds to an intake of 7.7 g of DF/day and 2.0 g of fructan/day. However, due to the low molecular weight of fructan, it is excluded in the DF assay by traditional methods (AOAC, Asp et al 1983). According

to the suggested new definition of DF (Anonymous 2001a), fructans are also included in DF. This means that the DF content of some cereal foods might be increased by as much as 20% due to the presence of fructans in the grain. The increase in soluble DF is still greater; in rye bran, the amount of soluble DF was increased from 4.5 to 11 g/100 g (Table III).

Rye fructans are water-soluble and easily retrieved in the water extract. The water-extractable concentrate of rye bran contained as much as 23 g of fructan/100 g. The three small fructo-oligosaccharides (DP 3–5) (Table III) were the only fructo-oligosaccharides that could be quantified because of availability of commercial standards. They made up 33% of the total fructan content in the water-extractable concentrate of rye bran. In whole grain, these three fructo-oligosaccharides formed 24% of the total fructan content. This is in good agreement with Henry and Saini (1989). The remainder of the fructan was apparently composed of longer oligosaccharides. The greater proportion (33%) of the three fructo-oligosaccharides of the total fructan content in water-extractable concentrate than in rye grain (24%) may be due to the processing of rye bran; fructan may have hydrolyzed during processing or only the smallest fructans had solubilized.

The amount of the three fructo-oligosaccharides was 1.2 g/100 g in rye grain and 7.4 g/100 g in the water-extractable concentrate of rye bran. Campbell et al (1997) and Hogarth et al (2000) have quantified fructo-oligosaccharides of rye grain and they found these small fructo-oligosaccharides at levels of 0.4–1.2 g/100 g, which is in the same range as the results of the current study.

Commercial inulin is a well-known prebiotic (Van Loo et al 1999) fermented quickly in vitro (Karpainen et al 2000). The fermentation pattern of cereal fructans has not been much studied, but these fructans may have the same prebiotic type properties as commercial inulin. In our preliminary studies (*unpublished results*), cereal fructans were fermented quickly during in vitro fermentation studies with human fecal inoculum.

## CONCLUSIONS

Rye is an important natural fructan source in human nutrition in countries where rye is consumed. Whole grain rye is also a good source of DF, especially after the suggested new definition of DF, which classifies fructans as DF. Fructan content of rye varies from year to year. Breadmaking may also diminish the fructan content. As a soluble carbohydrate, cereal fructans are quickly fermented by intestinal bacteria and they may have same health effects as commercial inulin.

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