Content of β-D-glucan in cereal grains

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Summary

The β-D-glucan content in 111 genotypes of spring barley, 79 genotypes of oat, and 14 genotypes of spring wheat was analyzed using the Mixed-linkage β-glucan assay kit. Samples were suspended and hydrated in a sodium phosphate buffer (pH 6.5), incubated with purified lichenase enzyme, and an aliquot of filtrate was reacted with purified β-glucosidase enzyme. The glucose product was assayed using an oxidase/peroxidase reagent. The mean value of β-D-glucan content in barley samples was 41.6 $g.kg^{-1}$ (in the range 18.6–53.7 $g.kg^{-1}$), in oats 34.9 $g.kg^{-1}$ (in the range 17.3–57.0 $g.kg^{-1}$), and in wheat samples 4.8 $g.kg^{-1}$ (in the range 1.9–6.7 $g.kg^{-1}$). Although we have observed high variation in β-D-glucan content within barley and oat genotypes, our results indicate that barley and oat are suitable sources of the health beneficial β-D-glucan. The barley genotypes Merkur, Orbit, Heda, Zlatan, Karát, Atlas, and Atribut, oat genotypes Terra, Avenuda, SV-5, and Arnold with the highest levels (> 50.0 $g.kg^{-1}$) observed can be used in our breeding programmes and in food industry as a potential material by preparation of functional foods.

Keywords

β-D-glucan; cereals; barley; oat; wheat

Whole grains became an ingredient of the human diet with the advent of agriculture about 10 000 years ago [1]. During the last 3000–4000 years, the majority of human world's population has relied upon whole grains as the main proportion of their diet. The positive effects of the whole grains and cereal products on human health have long been known. Yet in the 4th century BC, Hippocrates - the father of medicine - recognized the health benefits of the whole-grain bread. More recently, in the early 1800s to mid 1900s, physicians and scientists recommended whole grains to prevent constipation [2]. Whole grains are rich in nutrients and phytochemicals with known health benefits. They contain high amount of dietary fibre, resistant starch, oligosaccharides, antioxidants, micronutrients, phenolics, and other compounds related to disease prevention.

On 21 January 1997, the U. S. federal authority Food and Drug Administration (FDA) published health claim on food-product packages stating that "A diet high in soluble fibre from whole oats (rolled oats, oat bran, oatmeal, and oat flour) and low in saturated fat and cholesterol may reduce the risk of heart disease" [3, 4]. The beneficial effect of oat products is primary attributed to the soluble dietary fibre compound - (1→3)(1→4)-β-D-glucan, also called β-D-glucan [4, 5]. β-D-Glucan is a linear molecule of partially water-soluble polysaccharide consisting of glucose [6] linked by both β-(1→3) and β-(1→4)-linkages [7]. It is a major component of endosperm cell walls and subaleurone layer [8] of *Gramineae*, especially in oat (*Avena sativa* L.) and barley (*Hordeum vulgare* L.) [9-11]. It is also present at much lower levels in cell walls of rye (*Secale cereale* L.), maize (*Zea mays* L.), rice (*Oryza sativa* L.), sorghum (*Sorghum bicolor* L.), and millet (*Pennisetum americanum* L.) [12].

The activity of cereal β-D-glucan on the human and animal organisms has a broad range of effects. It has been shown that β-D-glucan from barley is hypocholesterolemic [13, 14], and this property may result of its ability to increase viscosity of intestinal content [15]. The recommended amount of fibre needed to effect cholesterol levels in humans

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is about 3 grams a day, to be taken as four times at least 0.75 grams per serving [4]. The hypocholesteromic effect of β-D-glucan is primarily related to its structure, molecular weight, and interaction with other components [16]. Further, β-D-glucan is a potent inductor of humoral and cell-mediated immunity, and regular daily consumption of β-D-glucan significantly increases immunological activity [17]. In vitro addition of oat β-D-glucan to cultivated macrophages increases production of interleukin IL-1 and tumor-necrosis factor TNF-α [17]. ESTRADA and co-workers [17] reported that an intraperitoneal administration of oat β-D-glucan to mice increased both the number and activity of peritoneal macrophages.

Furthermore, phagocytosis stimulation experiments showed that β-D-glucan is an important agent for maintenance of some blood biochemical parameters. It is responsible for decreasing total and LDL-cholesterol, ratio of total to HDL-cholesterol [18], and so it may reduce the risk of coronary heart disease and ischemic heart disease [19]. This type of dietary fibre can also modulate the glycaemic index [20] beneficial for patients with diabetes. As an active compound of soluble dietary fibre, β-D-glucan has an important role in gut physiology. Oatmeal's whole grain, high fibre, and protein attributes are believed to be the primary factors influencing spatial memory performance of children [21]. Moreover, oat's total and soluble fibre may contribute to favourable blood pressure parameters [22].

The physical and physiological properties of cereal β-D-glucan are of commercial and nutritional importance [23]. Increasing interests in cereal β-D-glucan during the last two decades are largely due to their acceptance as functional, bio-active ingredient [24]. OatLife (concentrated β-D-glucan made from oat bran) was the crucial ingredient in Primaliv, yoghurt and muesli combination, the first food labelled as "functional food" according to new rules in Sweden [25]. Based on its rheological characteristics, β-D-glucan has been also proposed to be applied as food hydrocolloid [26]. β-D-Glucan can be utilized as thickening agent to modify the texture and appearance in gravies, salad dressings, and ice cream formulations [9] or may be used as fat mimetic in the development of calorie-reduced foods [27]. Cereal β-D-glucan may also influence the sensorial quality of beverages [28]. Expansion in the functional drink sector has been tremendous over the last few years [29].

On the other hand, the excess of β-D-glucan influences brewing parameters of malt [30, 31]. It may cause a retarded extraction, lowering the yield in mashing as well as other problems with filtration during brewing because the dissolved β-D-glucan creates viscous solutions [32, 33]. Hence, in the malting process, the efficient degradation of the β-D-glucan from initial level of about 3.0–4.5% to about 0.2–1.0% is a critical factor [32].

It is clear, that the knowledge of β-D-glucan levels in cereal grains may be useful for consumers, and it would create an opportunity for scientists and breeders to develop new cultivars enriched in nutraceuticals like β-D-glucan. For example, researchers in Agriculture and Agri-Food Canada's Cereal Research Centre (Winnipeg) have developed the ELISA assay allowing plant breeders rapidly select individual plants possessing high level of β-D-glucan [25]. The aim of our study was to characterize three different sets of barley, oat, and wheat genotypes as suitable sources of β-D-glucan to be used in our breeding programs, and to consider selected cereals as a potential material for food industry.

MATERIAL AND METHODS

Seed samples of 111 spring barley (*Hordeum vulgare* L.) genotypes, 79 oat genotypes (*Avena sativa* L.), and 14 genotypes of wheat (*Triticum aestivum* L.), harvested in 2003, have been used in this study. The β-D-glucan content was analyzed in these samples as well as in some cereal-based commercial products, two sorts of peeled barleys and five sorts of oat flakes. Mature grains and commercial products were milled and passed through 0.5 mm sieve using Ultracentrifugal Mill (ZM 100 Retsch, Germany). The level of β-D-glucan was determined using Mixed-linkage β-glucan assay kit (Megazyme, Ireland) based on the method published by MCCLEARY and CODD [34]. This method is accepted by the AOAC (Method 995.16) and the AACC (Method 32-23) [35]. Samples were suspended and hydrated in a sodium phosphate buffer (pH 6.5), incubated with purified lichenase enzyme, and an aliquot of filtrate was reacted with purified β-glucosidase enzyme. The glucose product was assayed using an oxidase/peroxidase reagent. The measurement of the amount of β-D-glucan was performed in two (oat, wheat, commercial products) and three (barley) parallel assays, respectively. The values of the β-D-glucan levels were achieved for each sample as the mean ± standard deviation. The β-D-glucan content was calculated in dry weights.

Tab. 1. Content of β-D-glucan in barley genotypes.

No.	Barley genotype	The β-D-glucan content [g.kg^{-1}]	
		Mean value	SD
1	Nitran	18.6	0.4
2	Stabil	28.2	1.8
3	Stupický Hanácky	29.7	1.8
4	Expres	31.6	1.3
5	Krajová St. Hrozenkov	31.6	1.2
6	Terrasol Pivovarský	32.3	0.5
7	Zenit	33.3	1.2
8	Detěnický Kargyn	34.1	0.9
9	Spartan	34.8	0.9
10	Bonus	35.1	1.0
11	Forum	35.1	0.9
12	Viktor	35.2	0.7
13	Hanácký Kargyn	35.6	0.8
14	Kompakt	35.9	1.1
15	Hanácký exportný	36.0	0.3
16	Pudmerický pivovar	36.3	0.2
17	Čelechovický hanácký	36.6	0.8
18	Hanácký jubilejný	36.9	0.9
19	Fatran	37.2	0.1
20	Tolar	37.3	0.4
21	Hanácký Moravan	37.4	0.1
22	Dobrovický staročeský	37.6	0.8
23	Diosecký	37.7	0.9
24	Krajová z Orlové	37.8	0.3
25	Diamant	37.8	0.8
26	Galan	37.9	1.1
27	Janovický	37.9	1.5
28	Židlochovický	37.9	0.6
29	Amos	38.5	0.7
30	Diosecký 802	38.5	0.9
31	Hana	38.5	0.9
32	Progres	38.5	1.1
33	Sladko	38.5	0.8
34	Kosan	38.6	0.2
35	Novum	38.6	1.4
36	Bohatýr	38.9	0.8
37	Moravský nepolehavý IGB	39.0	0.9
38	Lutskij	39.0	0.7
39	Novodvorský Hanácky	39.0	0.8
40	Krystal	39.2	1.0
41	Dregerův	39.3	1.2
42	Valtický	39.3	0.8
43	Amulet	39.5	0.9
44	Ladik	39.6	0.8
45	Ametyst	39.8	0.9
46	Diosecký kneifel	39.9	0.5
47	Safír	39.9	0.6
48	Dukát	40.6	0.2
49	Dvoran	40.7	0.5
50	Branišovický výnosný	40.9	0.7
51	Nitriansky Exportný	40.9	1.1
52	Ekonom	41.1	1.0
53	Kvasický	41.2	0.9
54	Olbran	41.5	0.5
55	Export ratborský	41.6	0.9
56	Kredit 21	41.6	1.2
57	Granát	41.8	0.8
58	Rapid	42.1	1.0
59	Hanácky Staroveský	42.2	0.9
60	Víglašský Polojemný	42.2	1.1
61	Sladar	42.3	0.4
62	Diabas	42.4	0.6
63	Dregerův imperial	42.6	0.8
64	Garant	42.6	0.5
65	Stupický plnozrnný	42.6	0.7
66	Horal	42.9	0.6
67	Koral	43.4	0.8
68	Zefir	43.4	1.1
69	Akcent	43.6	0.8
70	Jarek	43.7	0.5
71	Svit	43.8	0.2
72	Tatry 1995	43.8	1.0
73	Šumavský	43.9	0.5
74	Zborovický Kargyn	43.9	0.8
75	Malvaz	44.2	0.7
76	Pisárecký	44.5	0.4
77	Proskovcuv	44.6	0.8
78	Heran	44.8	0.9
79	Favorit	44.9	0.8
80	Jindrichovický K64	44.9	0.7
81	Semčiský hospodársky	45.0	0.5
82	Rubin	45.3	1.0
83	Donum	45.5	0.4
84	Hořický	45.6	0.2
85	Semčiský pivovar	45.6	0.5
86	Jantar	45.8	0.8
87	Opál	45.8	0.8
88	Denar	45.9	0.4
89	Profit	46.0	1.5
90	Cyril	46.1	0.1
91	Olešenský	46.2	0.2
92	Slovenský Kvalitný	46.5	0.6
93	Terno	47.0	0.8
94	Moravský Nepoliehavý RIII/12	47.1	1.0
95	Jubilant	47.4	1.0
96	Nolč Dregeruv veleraný	47.7	0.7
97	Jaspis	47.8	0.4
98	Vladan	47.9	0.8
99	Topas	48.3	0.5
100	Michalovický	48.4	0.1
101	Ludan	48.7	0.5
102	Krukanický horský	49.2	1.2
103	Jarohnevický	49.4	0.8
104	Perun	49.9	0.7
105	Atribut	50.0	1.1
106	Atlas	50.1	0.7
107	Karát	51.7	0.8
108	Zlatan	52.1	1.0
109	Heda	52.7	0.8
110	Orbit	52.8	1.0
111	Merkur	53.7	1.7

SD - standard deviation.

RESULTS AND DISCUSSION

The mean values of β-D-glucan content in cereals were: spring barley 41.6 ± 0.6 g.kg^{-1}, oat 34.9 ± 0.8 g.kg^{-1}, and wheat 4.8 ± 0.2 g.kg^{-1}. Differences among genotypes in all cereal crops were observed.

The amount of β-D-glucan content in barley genotypes (Table 1) ranged between 18.6 g.kg^{-1} (genotype Nitran) and 53.7 g.kg^{-1} (Merkur). Previously published papers revealed that content of β-D-glucan in barley was 30.0–45.0 g.kg^{-1} [32] and up to 43.8 g.kg^{-1} [36]. In this study, we observed very high levels (> 50.0 g.kg^{-1}) of β-D-glucan in genotypes Merkur 53.7 ± 1.7 g.kg^{-1}, Orbit 52.8 ± 1.0 g.kg^{-1}, Heda 52.7 ± 0.8 g.kg^{-1}, Zlatan 52.1 ± 1.0 g.kg^{-1}, Karát 51.7 ± 0.8 g.kg^{-1}, Atlas 50.1 ± 0.7 g.kg^{-1}, and Atribut 50.0 ± 1.1 g.kg^{-1}. On the other hand, the lowest levels (< 30.0 g.kg^{-1}) were observed in genotypes Nitran 18.6 ± 0.4 g.kg^{-1}, Stabil 28.2 ± 1.8 g.kg^{-1}, and Stupický Hanácký 29.7 ± 1.8 g.kg^{-1}.

In contrast to barley, oat is less rich in β-D-glucan (Table 2). We have found that its mean content was 34.9 ± 7.9 g.kg^{-1}. REDAELLI et al. [37] reported the average amount of β-D-glucan in a set of 60 European oat genotypes to

Tab. 2. Content of β-D-glucan in oat genotypes.

No.	Oat genotype	The β-D-glucan content [g.kg^{-1}]	
		Mean value	SD
1	Edit	17.3	0.9
2	Fusch	20.0	0.4
3	Borka	20.2	0.4
4	PS 106	20.9	1.1
5	Vok	22.4	1.2
6	BE 201700	25.5	0.9
7	Autevil	25.9	0.4
8	INO 9801	26.6	0.5
9	Flamingstern	26.7	0.8
10	Petra	27.2	0.9
11	Flamingsprofi	27.4	1.1
12	Gambo	27.8	0.7
13	Orfine	27.9	0.2
14	Euro	28.4	0.4
15	German	28.5	0.5
16	Šampionka	28.9	0.2
17	Tarra	28.9	0.8
18	Lutz	29.1	0.9
19	Argentina	29.1	0.8
20	Kanton	29.3	1.0
21	Vilma	29.3	1.4
22	Dagny	29.5	0.8
23	10026 CN	30.0	0.2
24	Jumbo	30.0	0.1
25	Bug	30.6	0.1
26	Sanova AS 181325	31.0	0.8
27	Indio	31.7	0.1
28	Zlaťák	31.8	0.1
29	Auron	32.3	1.2
30	Aveia Peluda	32.9	0.7
31	Pan	33.1	0.8
32	Taiko	33.2	0.9
33	Expander	33.3	0.4
34	Noirine	33.5	0.5
35	Aragon	33.8	0.6
36	PS100	33.8	1.0
37	Leo	33.9	0.7
38	Ardo	33.9	0.8
39	Flipper	34.2	0.3
40	Flamingstrend	34.2	0.1
41	Sirene	34.2	0.5
42	Roxton	34.5	0.7
43	Selma	34.7	0.8
44	Pluco	34.9	0.5
45	Flamingslord	35.2	1.4
46	Consul	35.3	1.2
47	PS 90	35.6	1.0
48	Revisor	35.6	0.1
49	Maris Oberon	36.3	0.4
50	BE 202299	36.4	0.7
51	Azur	36.6	0.5
52	BE 211201g	36.8	0.1
53	Cyril	37.1	0.1
54	Master	37.1	0.5
55	Nelson	37.1	0.6
56	Senátor	37.2	1.1
57	Flamingsplus	37.3	0.7
58	Sevenanthree	38.3	1.1
59	Detvan	38.5	0.5
60	Adam	38.6	0.1
61	Stormont Sceptre	38.6	0.9
62	Avesta	38.7	0.7
63	Caracas	39.3	0.6
64	Zvolen	39.4	0.4
65	Dalimil	40.6	0.3
66	Ábel	40.7	0.8
67	Bandicoot	42.3	1.0
68	Poncho	44.0	1.4
69	Neklan	44.4	0.9
70	Pendek	45.4	0.4
71	Atego	45.6	0.4
72	Unisignum	45.9	0.4
73	Salomon	48.4	0.9
74	Izak	48.8	0.8
75	Neon	48.9	0.9
76	Arnold	52.0	0.9
77	SV-5	53.0	1.0
78	Avenuda	54.3	1.8
79	Terra	57.0	0.3

SD - standard deviation.

Tab. 3. Content of β-D-glucan in wheat genotypes.

No.	Wheat genotype	The β-D-glucan content [g.kg-1]	
		Mean value	SD
1	CDC Bounty	6.7	0.1
2	Timgalen	6.5	0.0
3	Lyall Pur	6.1	0.1
4	Landrace	6.0	0.1
5	CV. Alaska	6.0	0.5
6	Spalda (Landrace 1-96)	4.7	0.0
7	CV.TM 1	4.4	0.8
8	Landrace 1/99	4.3	0.1
9	CV.NO.48478	4.2	0.1
10	Blauer Santiger	3.9	0.1
11	Arcola	3.2	0.1
12	Kamut	3.0	0.1
13	CV.Einkorn	2.0	0.1
14	Local 1/99	1.9	0.1

SD - standard deviation.

Tab. 4. Content of β-D-glucan in oat flakes.

No.	Producer of the commercial product	The β-D-glucan content [g.kg-1]	
		Mean value	SD
1	Levice, Slovakia	26.4	0.1
2	Martin, Slovakia	32.1	6.0
3	Strančice, Czech Republic	36.1	1.4
4	Nemšová, Slovakia	42.8	0.3
5	Kráľová nad Váhom, Slovakia	46.0	1.8

SD - standard deviation.

Tab. 5. Content of β-D-glucan in peeled barley.

No.	Producer of the commercial product	The β-D-glucan content [g.kg-1]	
		Mean value	SD
1	Martin, Slovakia	33.5	0.6
2	Bratislava, Slovakia	37.4	0

SD - standard deviation.

be 38.9 g.kg^{-1}. In our study, very high level (> 50.0 g.kg^{-1}) of β-D-glucan was observed in genotypes Terra 57.0 ± 0.3 g.kg^{-1}, Avenuda 54.3 ± ± 1.8 g.kg^{-1}, SV-5 53.0 ± 1.0 g.kg^{-1}, and Arnold 52.0 ± 0.9 g.kg^{-1}. Very low content (< 20.0 g.kg^{-1}) was in genotypes Edit 17.3 ± 0.9 g.kg^{-1}, Fusch 20.0 ± ± 0.4 g.kg^{-1}, and Borka 20.2 ± 0.4 g.kg^{-1}.

Finally, the content of β-D-glucan in analyzed wheats was very low, ranging from 1.9 g.kg^{-1} to 6.7 g.kg^{-1} (Table 3). These results suggest that wheat is not a rich natural source of this health beneficial compound.

The β-D-glucan content was also analyzed in two and five barley- and oat-based commercial products, respectively (Table 4 and Table 5). The content of β-D-glucan ranged from 26.4 g.kg^{-1} to 46.0 g.kg^{-1} among five oat flakes. In peeled barleys, the content of β-D-glucan was 33.5 g.kg^{-1} to 37.4 g.kg^{-1}.

CONCLUSIONS

In this study, we presented a quantitative analysis of health-beneficial β-D-glucan in grains of the three most important cereal species: wheat, barley, and oat. The amount of β-D-glucan was determined by Mixed-linkage β-glucan assay procedure. The highest content of β-D-glucan was observed in barley and oat grains. The amount of β-D-glucan content in barley genotypes ranged between 18.6 g.kg^{-1} (genotype Nitran) and 53.7 g.kg^{-1} (Merkur), with the mean of 41.6 g.kg^{-1}. The mean value of β-D-glucan content in oat samples was 34.9 g.kg^{-1} with the range 17.3 g.kg^{-1} (genotype Fusch) and 57.0 g.kg^{-1} (genotype Terra). The barley genotypes Merkur, Orbit, Heda, Zlatan, Karát, Atlas, and Atribut and oat genotypes Terra, Avenuda, SV-5, and Arnold with the highest level (> 50.0 g.kg^{-1}) observed can be used in our breeding programmes as donors of health beneficial compound and in food industry as well as potential raw material for preparation of functional foods. Wheat was found unsuitable as source of the health beneficial β-D-glucan. The content of β-D-glucan in analyzed wheat genotypes ranged from 1.9 g.kg^{-1} to 6.7 g.kg^{-1}. We also determined the content of β-D-glucan in barley- and oat-based commercial products. The value of health beneficial fibre ranged from 26.4 g.kg^{-1} to 46.0 g.kg^{-1} in a group of five oat flakes. In peeled barleys, the content of β-D-glucan was 33.5 g.kg^{-1} to 37.4 g.kg^{-1}.

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