

Genotypic Variations in β -glucan Content of Barley Cultivated in Different Regions

Hong-Sik Kim*[†], Kwang-Geun Park*, Seong-Bum Baek*, and Jung-Hyun Nam*

*National Institute of Crop Science, RDA, Suwon 441-857, Korea

ABSTRACT: The level of β -glucan which is a major soluble dietary fiber found in the grain endosperm cell wall was highly variable among 25 barley genotypes grown at four locations including Suwon, Naju, Jinju, and Jeju. Statistically significant genotypic effects were observed for β -glucan content at each or across growing sites ($P < 0.001$). On average, 'Chalssalbori' showed the lowest percentage β -glucan (4.04%) among genotypes in the grain, whereas 'Yonezawa Mochi' was highest in percentage β -glucan (6.46%) compared to other genotypes. The significant difference between genotypes was approximately 1-2% across environments. The effects of location or interaction between locations and genotypes were not significant on the variation of β -glucan contents. High β -glucan content seemed to be greatly associated with such grain traits as waxiness and presence of husk except for 'Chalssalbori'. The waxy genotypes had a mean of 5.37% and values ranging from 5.28 to 5.47%, but normal genotypes had a mean of 4.78% and values ranging from 4.69 to 4.88% over environments. Hulless barley genotypes were also higher than hulled barley genotypes for the average β -glucan content in both individual and over all environments. The difference between the hulled and hulless gene pools was on average of 0.37% with ranges from 0.19% to 0.56% at four environments. β -glucan content measured from a mapping population of F_5 -derived 107 lines derived from the cross between 'Yonezawa Mochi' and 'Neulssalbori' was not significantly associated with other agronomic traits except for 1,000-kernel weight at the '01 Suwon environment. Not too much information on the relationship of β -glucan content to agronomic traits was available.

Keywords: barley, β -glucan, genotype, environment, variation, soluble dietary fiber

As most people concern about pursuing better life styles, the trend of food oriented towards health diets and physiological functionality has been emerged since 1990s. In particular, intake of soluble dietary fibers can be beneficial to treat life-style related diseases such as constipation, hemorrhoids, diverticulitis, and irritable bowel syndrome

(Kang & Song, 1997; Seog *et al.*, 2002).

High fiber diets are usually obtained from plant foods. Plants have various types and amount of fibers such as pectin, cellulose, hemicellulose, gum, and lignin. Their availability and physiological metabolism are different depending on the level of water-solubility. For example, water-soluble fibers of pectin and gum are found inside plant cells, and slow the passage of food through the intestines but do nothing to increase fecal bulk. On the other hand, water-insoluble fibers of cellulose, hemicellulose, and lignin are major components of cell wall, and facilitate the passage of food in the digestive tract (Heller & Hackler, 1977).

Barley is one of the starch crop species like rice and maize, and has become an important source of food and feed. Its grains also contain 8-15% of proteins, Vitamin B group, and other minerals. Recently, barley and oat were selected as one of the top 10 healthy foods for an alternative of dietary fibers (10 Foods that pack a wallop. Time January 21, 2002. Vol. 159 No. 3).

Barley and oat contain β -glucan, a major soluble fiber component mainly found in the grain endosperm cell walls. β -glucan is non-starch polysaccharide composed of three or four cellulosic (1 \rightarrow 4) group units joined by single (1 \rightarrow 3) linkages in structure (Woodward *et al.*, 1983). A few evidences from previous studies suggest that soluble β -glucan having the property of viscosity has a specific hypocholesterolemic effect to transport the precursors of cholesterol out of the body, and also helps lower blood pressure in hypertensive patients (Newman *et al.*, 1989; Ikegami *et al.*, 1996). On the other hand, barley β -glucan has also been detrimental against for beer brewing and poultry feeding (Newman *et al.*, 1987). Highly relative viscosity of β -glucan causes filtration and haze problems during the brewing process and reduced fat digestibility in poultry. Therefore, the effect and the value of β -glucan should be considered depending on the end-use product and processing of barley. Breeding of barley cultivars for particular uses would be greatly assisted by the information on the differences of genotypes and their interaction with environments for β -glucan content. We report the comparisons of the β -glucan content among barley genotypes and the effect of agronomic and environmental factors on the variation of β -glucan content.

[†]Corresponding author: (Phone) +82-31-290-6730 (E-mail) kimhongs@rda.go.kr <Received August 7, 2006>

MATERIALS AND METHODS

A total of 25 genotypes consisting of 9 hulled and 16 hull-less barley were evaluated for β -glucan content. These cultivars were planted in four locations including Suwon, Naju, Jinju, and Jeju. The trials were arranged in a randomized complete block design with two replications except for Jeju in which only one replication was applied. In addition, a population of F₅-derived 107 lines derived from the cross between a Japanese hull-less barley cultivar, 'Yonezawa Mochi' and a Korean hull-less barley cultivar, 'Neulssalbori'

was also evaluated for β -glucan content in two locations of Suwon and Jinju. Experimental plots were arranged in a randomized complete block design with two replications. Cultivation methods including the quantity of sowed seeds and fertilization followed the standard cultivation method for winter cereal crops recommended by Rural Development Administration (RDA) in Korea.

Measurement of β -glucan content was followed by McCleary method which was dependent on a streamlined enzymatic reactions and absolutely specific for mixed-linkage β -glucan (McCleary & Mugford, 1997). In our studies,

Table 1. Variation of total β -glucan content (%) in 25 barley genotypes grown at four different locations.

Genotype	Suwon	Jinju	Naju	Jeju	Mean	Var _{env}	Note [†]
Chalssalbori	4.10	4.80	4.20	3.04	4.04	0.54	N, wx
Neulssalbori	4.01	4.11	4.05	4.09	4.06	0.00	N, Wx
Milyanggutbori	3.92	4.03	4.11	4.20	4.06	0.01	C, Wx
Namhyangbori	4.24	4.02	4.24	4.61	4.28	0.06	C, Wx
Mirakbori	4.11	4.45	4.68	3.96	4.30	0.11	C, Wx
Suwon320	4.43	4.54	4.27	4.41	4.41	0.01	C, Wx
Nubet	4.42	5.11	4.78	4.48	4.70	0.10	N, Wx
Suwon321	4.25	5.16	4.95	4.77	4.78	0.15	N, wx
Suwon308	5.22	4.77	4.59	4.75	4.83	0.07	C, Wx
Olbori	5.12	4.71	4.54	5.16	4.88	0.09	C, Wx
Olssalbori	4.72	5.34	5.19	4.78	5.01	0.09	N, Wx
SB7803G...40-0-0	5.38	5.31	5.00	4.43	5.03	0.19	N, Wx
Suwon322	4.94	5.28	4.94	5.17	5.08	0.03	N, Wx
Suwon313	5.45	4.87	4.81	5.97	5.27	0.30	N, wx
Saechalssalbori	5.07	5.19	5.27	5.67	5.30	0.07	N, wx
Suwon310	5.06	5.37	5.19	5.65	5.31	0.07	C, Wx
Suwon302	5.23	5.03	5.60	5.69	5.39	0.10	N, wx
SB78618	5.36	5.26	5.73	5.55	5.47	0.04	N, wx
Suwon302-1	5.84	5.80	4.83	5.45	5.48	0.22	N, Wx
Daebaekbori	5.71	5.49	5.31	5.50	5.50	0.03	C, Wx
Duwonchapssalbori	5.49	6.68	4.85	5.09	5.53	0.66	N, wx
SB7803G...40-0-1	5.38	5.02	5.91	6.02	5.58	0.22	N, wx
SB901020	5.95	5.58	5.54	5.35	5.61	0.06	N, wx
Seodunchalbori	5.50	5.67	5.32	6.08	5.64	0.11	C, wx
Yonezawa Mochi	6.72	6.90	5.90	6.33	6.46	0.20	N, wx
Mean	5.02	5.14	4.95	5.05	5.04		
Maximum	6.72	6.90	5.91	6.33			
Minimum	3.92	4.02	4.05	3.04			
LSD (0.05)	1.10	0.82	0.96	-			
CV (%)	10.6	7.8	9.2	-			

[†]Note : C-Covered (Hulled), N-Naked (Hulless); Wx-Normal, wx-Waxy starch

we followed the analysis protocol provided by Megazyme™ kit (Megazyme International Ireland Ltd) for mixed linkage β -glucans. Approximately 0.5 gram of finely milled barley flour was prepared. Samples after suspension and hydration in 5.0 mL of sodium phosphate buffer solution (20 mM, pH 6.5) were incubated with purified lichenase enzyme (10 U). An aliquot of the filtrate is then reacted to completion with purified β -glucosidase enzyme (0.2 U). The glucose produced was assayed using a glucose oxidase/peroxidase reagent. The absorbance of each sample was measured at 510 nm.

RESULTS AND DISCUSSION

A total of 25 barley cultivars were evaluated for β -glucan content. Table 1 shows the mean and ranges of their values in each and over environments. Statistically significant genotypic effects were observed for β -glucan content at each or across growing sites ($P < 0.001$). On average, 'Chalssalbori' showed the lowest percentage β -glucan (4.04%) among genotypes in the grain, whereas 'Yonezawa Mochi' was highest in percentage β -glucan (6.46%). The significant difference between genotypes was approximately 1-2% across environments. The average β -glucan content of 25 genotypes for individual environments ranged from 4.95% in Naju to 5.14% in Jinju, and was 5.04% for the across environments (Table 1).

The high variability in β -glucan content suggests the existence of genetic diversity in β -glucan among the barley genotypes tested in this study. A few studies have been conducted to determine the β -glucan content of barley. However, direct comparison of genotypes might be difficult since the level of β -glucan was known to be various depending on genetic materials and growing environments. In most cases, the accessed barley genotypes showed a range of 3-7% for total β -glucan (Hockett *et al.*, 1987; Izydorczyk *et al.*, 2000). We also had a similar result in the ranges of total β -glucan with the previous studies in the literature.

It seemed that β -glucan content was highly associated with specific grain traits such as waxiness and presence of husk. As shown in Tables 1 & 2, the magnitude of difference between waxy and normal, or hulled and hulless cultivars varied without regard to their growing environments. The average β -glucan content of 11 waxy genotypes was always higher than that of 14 normal genotypes over all environments (Table 2). The waxy genotypes had a mean of 5.37% and values ranging from 5.28 to 5.47%, but normal genotypes had a mean of 4.78% and values ranging from 4.69 to 4.88% over environments. The relationship of waxy gene with the improvement of β -glucan content was evident with the comparisons of two sister lines derived from the same cross, such as SB7803G..40-0-0 (normal and hulless, 5.03%) and SB7803G..40-0-1 (waxy and hulless, 5.58%) in this study. Hulless barley genotypes were also higher than hulled barley genotypes for the average β -glucan content in both individual and over all environments. The difference between the hulled and hulless gene pools was on average of 0.37% with ranges from 0.19% (Jeju) to 0.56% (Jinju) at four environments.

These results in our studies were congruent with the previous reports for the effect of waxy genotype and hullessness. Ullrich *et al.* (1986) reported that the mean β -glucan content of waxy isotypes in barley was 6.4%, which was higher than that (4.8%) of the normal isotypes. Izydorczyk *et al.* (2000) also observed significant differences in total β -glucan among the groups of high amylose, waxy, zero amylose waxy, and normal barley with average values of 7.49%, 6.86%, 6.30%, and 4.38%, respectively. However, along with a relatively poor correlation ($r=0.41$) between the soluble and total β -glucan, they suggested that relative rankings of solubility of β -glucans among these four groups might be changed in other studies due to different extraction parameters and methodologies. With regard to quality considerations for hulless barley, Hickling (1999) and Jadhav *et al.* (1998) reported that hulless barley had higher levels of β -glucans than covered barley since β -glucans were concen-

Table 2. Variation of average total β -glucan content (%) in the groups of different grain characteristics.

Grain characteristics	No. of lines	Suwon	Jinju	Naju	Jeju	Mean
Hulled	9	4.81	4.78	4.69	4.92	4.80
Hulless	16	5.14	5.34	5.09	5.12	5.17
Normal	14	4.79	4.88	4.69	4.76	4.78
Waxy	11	5.32	5.47	5.28	5.41	5.37
Hulled/Normal	8	4.72	4.67	4.61	4.78	4.70
Hulled/Waxy	1	5.50	5.67	5.32	6.08	5.64
Hulless/Normal	6	4.88	5.15	4.80	4.73	4.89
Hulless/Waxy	10	5.30	5.45	5.27	5.35	5.34

Table 3. Correlation between β -glucan content and agronomic traits for a population of F_5 -derived 107 lines derived from the cross between 'Yonezawa Mochi' and 'Neulssalbori' in the three environments.

Environment	Heading date	Maturity Period	Plant height	Head length	1000-kernel weight
Suwon '01	-0.06	0.15	0.05	0.07	0.32**
Suwon '02	-0.07	0.05	0.002	0.02	0.05
Jinju '02	0.09	-	-0.14	0.04	0.16

**Statistically significant at $P < 0.01$.

trated in the endosperm.

Most traits are genetically associated with the chromosomes. It is known that the genes controlling waxy endosperm (*wx*) and naked caryopsis (*n*) are located on the same chromosome 7H in barley (Hockett & Nilan, 1985). According to our genetic mapping studies, one of the major quantitative trait loci associated with the β -glucan content in 'Yonezawa Mochi' was found near *wx* gene on the chromosome 7H (data not shown, manuscript in preparation). This fact explains that the level of β -glucan content might be related by the presence of these traits due to their genetic linkages. As shown in our results, most of waxy and hulless barley genotypes showed a marked trend of increase of β -glucan level except for 'Chalssalbori'.

As shown in Table 3, β -glucan content measured from a mapping population of F_5 -derived 107 lines derived from the cross between 'Yonezawa Mochi' and 'Neulssalbori' was not significantly associated with other agronomic traits except for 1,000-kernel weight at the '01 Suwon environment. Not too much information on the relationship of β -glucan content to agronomic traits was available. Prentice *et al.* (1980) reported that barley genotypes with higher grain weight showed greater β -glucan content. Their results might be related with Hockett *et al.* (1987)'s study in which the 6-rows were consistently lower than the 2-rows in the β -glucan content. On the other hand, no clear relationship was found between the β -glucan content and kernel weight in the Izydorczyk *et al.*'s results (2000).

It is not clear how much the environmental factors influence the total β -glucan content. Ullrich *et al.* (1986) found that barley grown in the hot dry conditions in the grain-filling period had enhanced β -glucan content. On the other hand, the effect of temperature in the period from post tillering to flowering and soft dough development stage was significant and negatively correlated with β -glucan content (Hockett *et al.*, 1987). The relationship of moisture availability for plants and grain β -glucan content was not clear yet. Bourne & Wheeler (1984) and Coles (1979) reported strong association between moisture availability and β -glucan content, but not in the result of Hockett *et al.* (1987). Excessive moisture in the final stages of grain maturation made against the level of β -glucan (Stuart *et al.*, 1988).

Our results apparently revealed that total β -glucan content was more dependant on the genotypes than the environmental effects. This was consistent with the conclusion of Stuart *et al.* (1988). The effects of location or interaction between locations and genotypes were not significant on the variation of β -glucan contents among 25 barley genotypes grown at Suwon, Jinju, and Naju (data not shown). As shown in Table 1, most genotypes exhibited stable β -glucan level without regard to their growing environments. In particular, 'SB901020' and 'Daebaekbori' showed relatively high β -glucan level with smaller variation among environments. On the other hand, such genotypes with high β -glucan as 'Yonezawa Mochi' and 'Duwonchapsalbori' showed relatively greater variation than others.

Based on our current understanding of human food requirements, our breeding priorities should include hulless, waxy starch, high β -glucan, good cooking and pearling qualities in barley. Screening for germplasm with waxy starch and high β -glucan is commonly performed in the early stages of breeding selection. In addition, it should be clarified how the grain starch compositions such as amylose-free and waxy traits are related with the change of β -glucan content. Barley β -glucan has been found to contribute health benefits for people, but is a troublesome polysaccharide for making beer, and also for making fuel ethanol. Since, great variability can exist in total amount and degree of solubility of β -glucan in barley, it is important to develop ways to maximize this grain component for the various end-uses and by-products.

REFERENCES

- Bourne, D.T. and R.E. Wheeler. 1984. Environmental and varietal differences in total beta-glucan contents of barley and the effectiveness of its breakdown under different malting conditions. *J. Inst. Brew.* 90 : 306-310.
- Coles, G.D. 1979. Relationship of mixed-link beta-glucan accumulation to accumulation of free sugars and other glucans in the developing barley endosperm. *Carlsberg Research Communications* 44 : 439-453.
- Heller, S.N. and L.R. Hackler. 1977. Water-holding capacity of various sources of plant fiber [Foods]. *J. of Food Sci.* 42 : 1137-1143.

- Hickling D.R. 1999. Does the feed industry have barley quality needs Proceedings of Canadian Barley Symposium '99 pp. 55-58.
- Hockett, E.A. and Nilan, R.A. 1985. Genetics. In Barley (ed. D. Rasmusson), 187-230. Agron. Monog. No. 26. Am. Soc. Agron, Madison, U.S.A.
- Hockett, E.A., C.F. McGuire, and C.W. Newman. 1987. The relationship of barley beta-glucan content to agronomic and quality characteristics. Barley Genet. V : 851-860.
- Ikegami, S., M. Tomita, S. Honda, M. Yamaguchi, R. Mizukawa, Y. Suzuki, M. Higuchi, and S. Kobayashi, 1996. Effect of boiled barley-rice-feeding in hypercholesterolemic and normolipemic subjects. Plant Foods for Human Nutrition 49 : 307-316.
- Izydorczyk, M.S., J. Storsley, D. Labossiere, A.W. MacGregor, and B.G. Rossnagel. 2000. Variation in total and soluble β -glucan content in hullless barley: Effects of thermal, physical, and enzymic treatments. J. Agric. Food Chem. 48 : 982-989.
- Jadhav, S.J., S.E. Lutz, V.M. Ghorpade, and D.K. Salunkhe. 1998. Barley: Chemistry and value-added processing. Crit. Rev. Food Sci. 2 : 123-171.
- Kang, H.J. and Y.S. Song. 1997. Review : Dietary fiber and cholesterol metabolism. Korean J. Food & Nutr. 26 : 358-371.
- McCleary, B. V. and D.C. Mugford. 1997. Determination of beta-glucan in barley and oats by streamlined enzymic method: summary of collaborative study. J. Assoc. Off. Anal. Chem. 80 : 580-583.
- Newman, R.K., C.W. Newman, J. Fadel, and H. Graham. 1987. Nutritional implications of beta-glucans in barley. Barley Genetics V : 773-780.
- Newman, R.K., C.W. Newman, and H. Graham. 1989. The hypocholesterolemic function of barley β -glucan. Cereal Foods World 34 : 883-886.
- Prentice, N., S. Babler, and S. Faber. 1980. Enzymic analysis of beta-D-glucans in cereal grains. Cereal Chem. 57 : 198-202.
- Seog, H.M., S.R. Kim, H.D. Choi, and H.M. Kim. 2002. Effects of β -Glucan-enriched barley fraction on the lipid and cholesterol contents of plasma and feces in rat. Korean J. Food Sci. Technol. 34: 678-683.
- Stuart, I.M., L. Loi, and G.B. Fincher. 1988. Varietal and environmental variations in (1 \rightarrow 3, 1 \rightarrow 4)- β -Glucanase potential in barley: Relationships to malting quality. J. of Cereal Sci. 7 : 61-71.
- Ullrich, S.E., J.A. Clancy, R.F. Eslick, and R.C.M. Lance. 1986. Beta-glucan content and viscosity of extracts from waxy barley. J. Cereal Sci. 4 : 279-285.
- Woodward, J. R., D. R. Phillips, and G. B. Fincher. 1983. Water-soluble (1 \rightarrow 3),(1 \rightarrow 4)- β -D-glucans from barley (*Hordeum vulgare*) endosperm. I. Physicochemical properties. Carbohydr. Polymers 3 : 143-156.