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ARTICLE

Relationship between the Contents of Bioactive Components in Grain and the Release Dates of Wheat Lines in the HEALTHGRAIN Diversity Screen

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The EU FP6 HEALTHGRAIN has generated an extensive database on the contents of phytochemicals (alkylresorcinols, tocols, sterols, phenolic acids, folates) and dietary fiber components in the grain of wheat, including analyses of 150 lines grown on a single site in Hungary in 2005 and a smaller set of lines grown under five (three lines) or six (23 lines) different environments (in Hungary in 2005, 2006, and 2007 and in France, Poland, and the United Kingdom in 2007). The lines analyzed included land races and varieties bred between the mid-19th and early 21st centuries. These results have been analyzed to determine whether the contents of these groups of bioactive components in the grain have decreased with the development of intensive plant breeding in the second part of the 20th century. No decreases in the contents of any groups of bioactive components were observed in relation to release date, showing that selection for increased yield and protein quality has been effectively neutral for other grain components.

KEYWORDS: Wheat; dietary fiber; intensive breeding; bioactives; health benefits; phytochemicals

INTRODUCTION

Three cereal crops, wheat, maize, and rice, dominate world agricultural production, with total annual yields of about 2000 × 10^6 tonnes. However, wheat is the most widely grown, from 67° N in Scandinavia to 45° S in Argentina (*1*) and is also the most widely consumed by humans, after processing to give bread, pasta, noodles, cakes, cookies, and many other products. Consequently, it makes a significant contribution to the nutritional requirements of humans, even in developed countries such as the United Kingdom, where it is an important source of minerals (including Se, Mo, Fe, Zn), vitamins (niacin, thiamin, and vitamins B_6 and E), and dietary fiber (*2*) as well as calories (energy) derived from the high content of starch and protein.

However, it has also been suggested that intensive wheat breeding has resulted in decreased health benefits, by focusing on yield and processing quality. Sands and co-workers (*3*, *4*) have specifically suggested that intensive breeding has resulted in increases in the proportion of amylopectin in wheat starch and in the content of gluten proteins, due to the emphasis on selecting for baking and processing performance. Because amylopectin is more readily digested than amylose, such an increase could contribute to the development of insulin insensitivity and type 2 diabetes (*5*). The same authors also pointed out the role of gluten in triggering celiac disease, an autoimmune condition that is increasing in prevalence in many countries (*6*−*9*). The gluten proteins also have low contents of essential amino acids, notably lysine, compared with other grain proteins, so increases in gluten protein content are associated with reduced protein nutritional quality (reviewed in ref *10*). Reduced contents of essential minerals, including iron and zinc, have also been reported in modern high-yielding cultivars of wheat compared to older varieties (*11*−*14*).

The EU FP6 HEALTHGRAIN program (2005−2010) aimed to improve the health and well-being of consumers by increasing the intake of protective components present in wholegrain cereals (*15*). This included an extensive "diversity screen" of the extent of variation in the contents of selected bioactive components in 150 wheat varieties (*16*). This was followed by more detailed analyses of 26 varieties grown in six site × year combinations to determine the contributions of genotype and environment and the heritabilities of the components (*17*). These analyses showed that the content of zinc, and possibly to a lesser extent

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Table 1. Lines with High and Low Contents of Phytochemicals and Dietary Fiber Components

lines with highest phytochemical score (26−29)		lines with lowest phytochemical score (7−10)		lines with highest fiber score (23−29)		lines with lowest fiber score (6−12)	
cultivar	release date	cultivar	release date	cultivar	release date	cultivar	release date
Estica	1990	San Pastore	1940	Lynx	1992	Frederick	1971
Monopol	1975	Aurora	1972	Yumai 34	1988	Glenlea	1972
Campari	2003	Spartanka	1988	Maris-Huntsman	1971	Buck Catriel	1992
Malacca	1997	Nap Hal		Sagittorio	1994	Red River 68	1968
Kanzler	1980	Gloria	1977	Recital	1986	Key	1976
Augusta	1979	Skorospelka 3B	1955	Milan	1988	Sultan 95	1995
Thatcher	1934	Balkan	1979	Campari	2003	Chara	1998
Atlas 66	1948	Agron	1980	NS Rana 1	1975	Thatcher	1934
Moulin	1985	Sadovo-	1972	Moulin	1985	Monopol	1975
Hereward	1989	Alabasskaja	1947	Amadeus	1985	Kanzler	1980
Lynx	1992	Bezostaya	1911	Gloria	1977	Korweta	1997
Cadenza	1992	Krasnodarskaya 99	2003	Valoris	1998	San Pastore	1940
Claire	1999	Sava	1967	Balkan	1979	Baranjka	1979
Aktuer	2004	Gerek 79	1979	Lasta	1987	Alabasskaja	1947
Rialto	1993	Autonomia	1938	Tam 200	1986	Alba	1987
Disponent	1975	Obrii	1983	Seu Seun 27	1936	Isengrain	1997
Tremie	1992	Albatros Odeskii	1990	MV Suba	2002	Riband	1987
Ellvis	2002	Catbird	1991	Magdalena	1949	Akteur	2004
Tommi	2002			Ble des Domes	1940	Lona	1991
Riband	1987			Momtchil	1982	Qualital	1991
				Sadovo 1	1972	Soissons	1987
				Zvezda	1982		

also of iron, decreased with release date, which was probably associated with the introduction of modern semidwarf varieties (as reported in ref *14*). By contrast, the analysis reported here shows that there has been no decline in the contents of other bioactive components, refuting suggestions that the health benefits of wheat have decreased significantly due to intensive plant breeding.

MATERIALS AND METHODS

One hundred and fifty lines of bread wheat (*Triticum aestivum* var. *aestivum*) (listed in **Table 1** (taken from ref *16*) were selected from Europe, Asia, the Americas, and Australia to include land races and breeding lines as well as modern and older cultivars. One hundred and thirty of the lines were of winter type, whereas 20 were spring type, and they included representatives of all of the major end-use categories, with soft or hard texture, red or white color, and low or high protein content. The lines were sown in small plots (6 rows, about 2.5 × 1.2 m) at the Agricultural Research Institute of the Hungarian Academy of Sciences, Martonvásár, Hungary, in 2004 (winter types) or 2005 (spring types) as described in ref *16*. Because of the small plot size, which would result in significant edge effects, it is not valid to convert the plot yields (expressed as kg/plot) to tonnes per hectare.

From the 150 bread wheat lines tested in the diversity screen, 23 lines were selected to represent a range of contents of phytochemicals (sterols, tocols, alkylresorcinols, folates, phenolic acids) and dietary fiber components, as described in ref *16*. They were then grown together with the control cultivar, MV Emese, and two additional lines (Tiger and Crousty) at the same site in Martonvásár in 2006 and at four sites in 2007 (Martonvásár, Hungary; Woolpit, U.K.; Choryn, Poland; Clermont Ferrand, France) (see ref *17* for details).

Agronomic treatments were standard for the individual sites, with 110 kg of N/ha being applied in Poland, 204 kg of N/ha in the United Kingdom, 200 kg of N/ha in France, and 140 kg of N/ha in Hungary, and the appropriate use of agrochemicals.

Milling was carried out using a Perten Laboratory Mill 3100 (with a 0.5 mm sieve) to produce wholemeal and with a Chopin CD1 laboratory mill to produce bran and flour. Milled samples were rapidly cooled to −20 °C for storage. Analysis of bioactive components was carried out as described previously (*18*−*23*), using wholemeal for all components except arabinoxylan fractions (AX), which were determined on white flour and bran.

To calculate total scores for phytochemicals and dietary fiber components, the lines were grouped into six classes for each phytochemical and dietary fiber component, with group 6 comprising the 25 lines with the highest contents and group 1 the 25 lines with the lowest contents. Addition of the scores for the individual phytochemicals means that lines with the highest levels of all components (tocols, sterols, alkylresorcinols, phenolic acids, folates) would have a maximum score of 30. Similarly, lines with the highest levels of all dietary fiber (DF) components (wholemeal β-glucan percent, flour WE-AX percent, flour total AX percent, bran WE-AX percent, bran total AX percent) would also have a maximum score of 30.

RESULTS

Release dates were available for 146 of the 153 lines included in the HEALTHGRAIN study; two of the lines (Nap Hal and Chinese Spring) were known to be land races, which were not commercially released. These 148 lines are therefore listed in Table S1 in the Supporting Information, in which the 123 lines grown in 2005 only are shown in lower case, the 22 lines grown in 2005−2007 in capitals, and the three lines grown in 2006−2007 only in italicized capitals. The relationships between the contents of bioactive components and release dates were then determined for the samples grown only in 2005 and for those grown in 2006 and in the multisite trial in 2007.

Analysis of Lines Grown in 2005. To display the results graphically, an arbitrary date of 1900 was used for the two land races (Nap Hal and Chinese Spring) and for Red Fife, which was released in 1842. Of the remaining lines, 20 were released before 1960, 34 between 1960 and 1979, 36 between 1980 and 1989, 36 between 1990 and 1999, and 19 between 2000 and 2004. The lines were selected to reflect diversity in ancestry and geographical origin as well as release date, as discussed in ref *16*.

The limited availability of grain for some lines meant that only single small plots (6 rows, about 2.5 × 1.2 m) rather than large replicate plots, which are usually considered to be appropriate for yield determination, were available. Furthermore, many of the lines were grown outside their areas of adaptation and, hence, may have given atypically low yields. Therefore, although many of the varieties released after 1950 showed the expected increases in yield, the other varieties gave low yields, which probably resulted from poor adaptation to the south-continental climate

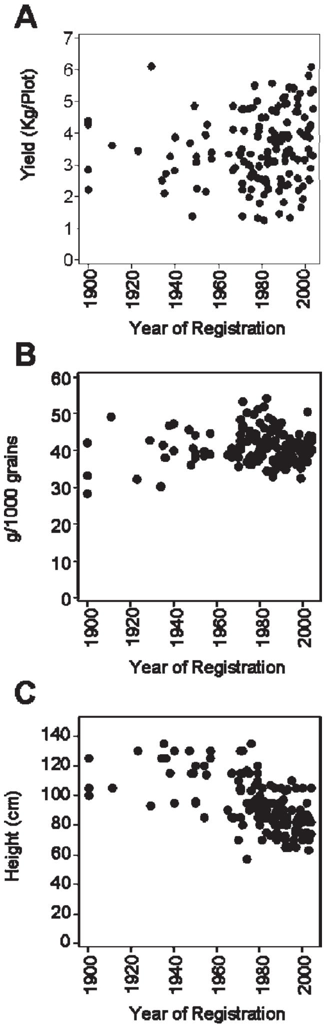

Figure 1. Relationship between release date of the 145 wheat lines grown in 2005 and yield (**A**), thousand grain weight (**B**), and plant height (**C**). The two land races (Chinese Spring and Nap Hal) and Red Fife (released 1842) are shown with release dates of 1900.

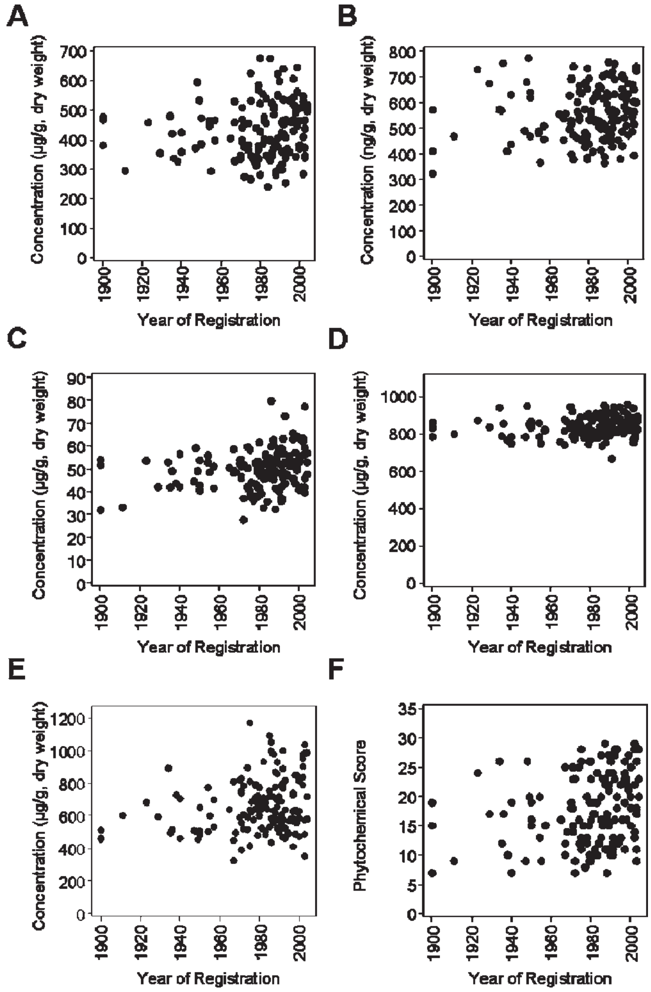

Figure 2. Relationship between release date and contents of phytochemicals for the 145 wheat lines grown in 2005: (**A**) alkylresorcinols; (**B**) folate; (**C**) tocols; (**D**) sterols; (**E**) total phenolic acids; (**F**) total phytochemical score. The two land races (Chinese Spring and Nap Hal) and Red Fife (released 1842) are shown with release dates of 1900.

in Hungary (**Figure 1A**). The thousand grain weights ranged from about 30 to 50 g (**Figure 1B**) with no relationship to date of release. Although this may partially reflect the fact that many of the lines were grown outside their range of adaptation, it is also consistent with the suggestion that increases in grain yield due to breeding are usually associated with increased seed number rather than weight (*24*). A decrease in the heights of the lines has occurred since the 1960s (**Figure 1C**), reflecting the widespread exploitation of dwarfing genes to reduce stem height.

Figure 2 shows the total contents of five major groups of bioactive phytochemicals in wholemeal flour, alkylresorcinols, folates, tocols, sterols, and phenolic acids, plotted in relation to release date. No relationships between release date and the contents of sterols (**Figure 2D**) or folates (**Figure 2B**) were observed, with the former being remarkably constant in content between the lines at 670−959 μg/g dry weight (as reported previously in ref *18*) and the latter more variable (364−774 ng/g dry weight) (*21*). By contrast, the range of variation in the contents of the other groups (alkylresorcinols, tocols, phenolic acids) (**Figure 2A**,**C**,**E**) increased with release dates, with some of the more recent lines having higher contents than the older lines. For example, the content of tocols ranged from 32 to 59 μg/g dry weight with an average content of 47 μg/g dry weight for the lines released up to 1959 and from 28 to 80 μg/g dry weight with an average content of 50 μg/g dry weight for the lines released between 1960 and 2004. Corresponding ranges for alkylresorcinols were 295−595 μg/g dry weight (average 419 μg/g dry weight) for the lines released up to 1959 and 241−676 μg/g dry weight (average 443 μg/g dry weight) for the lines released from 1960 to 2004. The total phenolic acid concentrations ranged from 456 to 892 μg/g dry weight (average 584 μg/g dry weight) for the lines released up to 1959 and from 326 to 1171 μg/g dry weight (average 679 μg/g dry weight) for the lines released from 1960 to 2004 (see refs *19*, *20*, and *22* for full data sets).

The phenolic acids were extracted as three fractions: free phenolic acids, which typically account for 0.5−1% of the total; soluble conjugated forms, which typically account for about 22%; and bound forms, which typically account for about 77% (*16*). Similar analyses of the amounts of these fractions in relation to release date (**Figure 3**) showed that many recent lines had much higher content of free phenolic acids than the older lines, with similar but smaller differences in the contents of conjugated and bound phenolic acids.

To compare the total contents of phytochemicals, the lines were assigned a "phytochemical score", based on their rankings (1−6) for the five groups of components (see Materials and Methods), which showed no decrease with release date (**Figure 2F**).

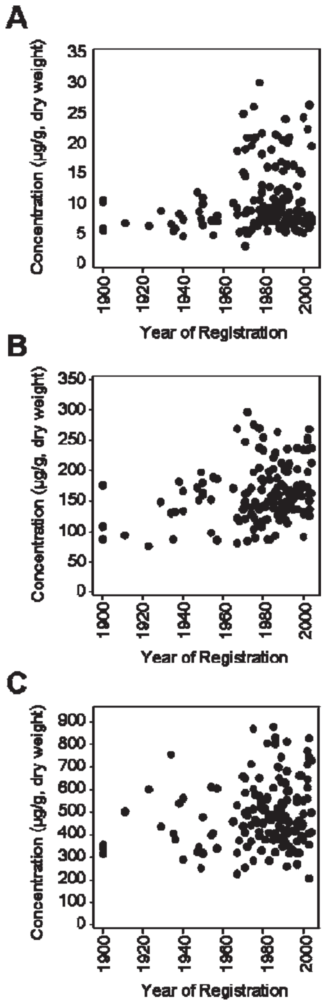

Figure 3. Relationship between release date and contents of phenolic acids for the 145 wheat lines grown in 2005: (**A**) total free; (**B**) total conjugated; (**C**) total bound. The two land races (Chinese Spring and Nap Hal) and Red Fife (released 1842) are shown with release dates of 1900.

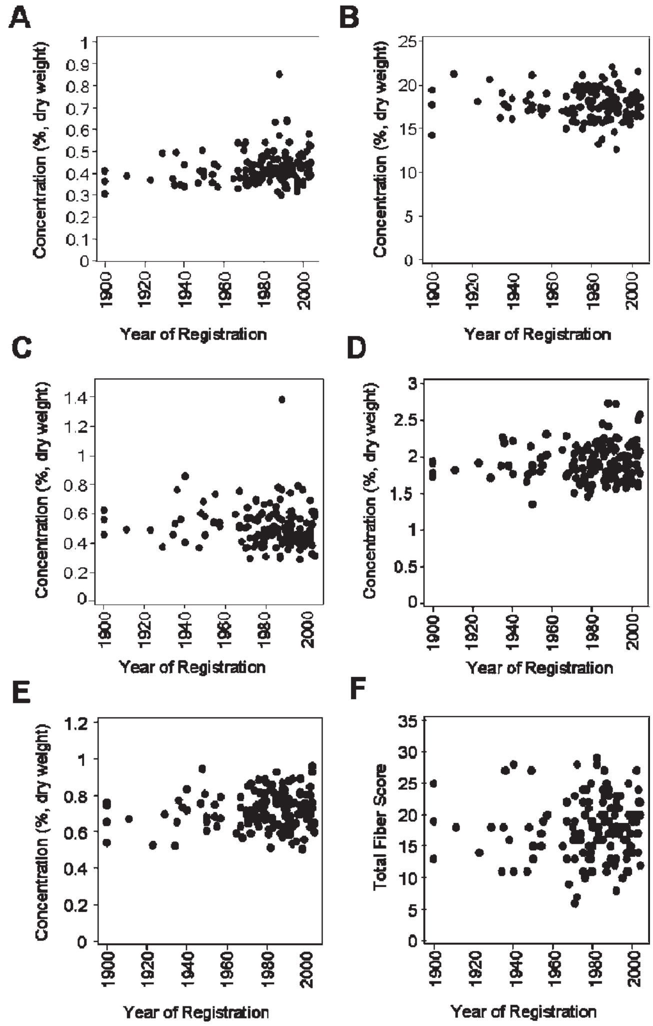

Figure 4. Relationship between release date and contents of dietary fiber components for the 145 wheat lines grown in 2005: (**A**) bran WE-AX; (**B**) bran TOT-AX; (**C**) flour WE-AX; (**D**) flour TOT-AX; (**E**) wholemeal β-glucan; (**F**) dietary fiber score. The two land races (Chinese Spring and Nap Hal) and Red Fife (released 1842) are shown with release dates of 1900.

Similar analyses of the contents of dietary fiber components against release date are shown in **Figure 4**, with water-extractable (WE) arabinoxylan (AX) and total (TOT) AX being determined in bran and white flour fractions and β-glucan in wholemeal flour. No relationship between release date is observed, with the "total fiber ranking" (**Figure 4F**) showing that the overall range is slightly greater for the modern lines than for the older lines.

The total phytochemical score and total dietary fiber score are plotted against grain yield in **Figure 5**, panels **A** and **C**, respectively, and against thousand grain weight in **Figure 5**, panels **B** and **D**, respectively. A weak negative correlation was observed between thousand grain weight and total phytochemical index ($r = -0.389$, $p < 0.0001$) (**Figure 5B**), which is consistent with previous analyses of the same data set that showed negative correlations between thousand grain weight and total tocols, total sterols, and total alkylresorcinols (*16*). These negative correlations probably result from the concentration of these components in the outer layers (bran) and germ, which account for a higher proportion of the total weight of small grain than of larger grain. By contrast, a weak positive correlation was observed between thousand grain weight and total dietary fiber index ($r = 0.312$, $p = 0.00014$) (**Figure 5D**), which is consistent with the previous report of a weak positive correlation between thousand grain weight and TOT-AX in bran (*16*).

Analysis of the Lines Grown in Multisite Trials 2005−2007. The 23 lines gown in 2005, 2006, and 2007 and the three additional lines grown only in 2006 and 2007 are shown in capitals and in italicized capitals, respectively, in Table S1 in the Supporting Information. In this case the mean contents of the various bioactive components for samples grown on the five/six sites (see refs *28*−*33* for full data sets) were plotted against release date (Figures S1 and S2 in the Supporting Information), confirming that the contents are not lower in recent compared to older lines.

DISCUSSION

The selected lines included land races, old varieties dating from 1842, and modern varieties released up to 2004. There have clearly been major changes in agronomic practices during this period, particularly increased use of fertilizers and crop protection chemicals since the middle of the 20th century. Furthermore, agronomic practices also vary between the countries of origin, with varieties being selected and grown under higher input conditions in western Europe than in most other regions. The agronomic regimens used to produce the HEALTHGRAIN samples were typical for the various countries, with higher levels of fertilizer N being applied in

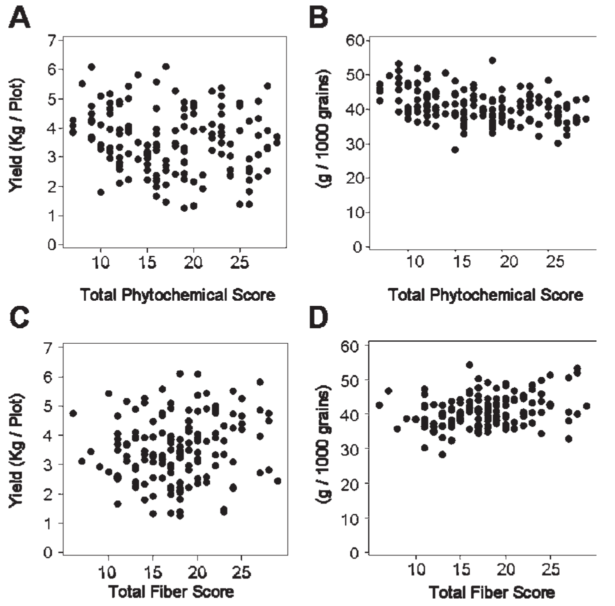

Figure 5. Relationship between total phytochemical score (**A**, **B**) and total dietary fiber score (**C**, **D**) with grain yield (**A**, **C**) and thousand grain weight (**C**, **D**) of the wheat lines grown in 2005.

the United Kingdom (204 kg/ha) and France (200 kg/ha) than in Hungary (140 kg/ha) and Poland (110 kg/ha) and appropriate use of other agrochemicals.

The analyses described here show no decline in the contents of bioactive components in wheat with release date, despite the fact that previous analyses of the same material showed decreased content of minerals since the introduction of semidwarf varieties (*25*). Negative correlations between grain yield and trace element concentrations have also been reported in some trials (*11*, *12*, *26*, *27*), but not in others (*34*).

Most of the 20 lines with the highest scores for phytochemicals had release dates after 1960, the exceptions being Thatcher (1934) and Atlas 66 (1948). The 18 lines with the lowest score for total phytochemical content contained lines with both pre- and post-1960 release dates. A similar observation was also made when scores for total dietary fiber components were analyzed. Of the 22 lines having the highest score for total dietary fiber content, 19 had release dates after 1960, with only 3 lines (Seu Seun 27, 1936; Magdalena, 1949; and Ble des Domes, 1940) being released up to 1959. The only lines with high scores for both phytochemicals and dietary fiber components were Lynx, Campari, and Moulin, which have release dates of 1992, 2003, and 1985, respectively.

Finally, it should be emphasized that the present study analyzed only a limited number of varieties, with most being grown on a single site for one year. More detailed studies on multiple with precise determination of yield components as well as bioactive components are therefore required to confirm the conclusions.

ABBREVIATIONS USED

AX, arabinoxylan; DF, dietary fiber; TOT-AX, total arabinoxylan; WE-AX, water-extractable arabinoxylan.

Supporting Information Available: Table S1 and Figures S1 and S2. This material is available free of charge via the Internet at http://pubs.acs.org.

LITERATURE CITED

(1) Feldman, M. Wheats. In *Evolution of Crop Plants*; Smartt, J., Simmonds, N. W., Eds.; Longman Scientific and Technical: Harlow, U. K., 1995; pp 185−192.
(2) Buttriss, J. Introduction: plant foods and health. In *Plants: Diet and Health*; Goldberg, G., Ed.; Blackwell Publishing: Oxford, U.K., 2003; pp 1−26.
(3) Morris, C. E.; Sands, D. C. The breeder's dilemma − yield or nutrition? *Nat. Biotechnol.* **2003**, *24*, 1078−1080.
(4) Sands, D. C.; Morris, C. E.; Dratz, E. A.; Pilgeram, A. L. Elevating optimal human nutrition to a central goal of plant breeding and production of plant-based foods. *Plant Sci.* **2009**, *177*, 377−389.
(5) Byrnes, S. E.; Brand Miller, J. C.; Denyer, G. S. Amylopectin starch promotes the development of insulin resistance in rats. *J. Nutr.* **1995**, *125*, 1430−1437.
(6) Lohi, S.; Mustalahti, K.; Kaukinen, K.; Laurila, K.; Collin, P.; Rissanen, H.; Lohi, O.; Bravi, E.; Gasparin, M.; Reunanen, A.; Mäki, M. Increasing prevalence of coeliac disease over time. *Aliment. Pharmacol. Ther.* **2007**, *26*, 217−225.
(7) Green, P. H. R.; Cellier, M. D. B. Celiac disease. *NEJM* **2007**, *357*, 1731−1743.
(8) Rodrigo, L. Celiac disease. *World J. Gastroenterol.* **2006**, *12*, 6585−6593.
(9) Fasano, A. Celiac disease: the past, the present, the future. *Pediatrics* **2001**, *107*, 768−880.
(10) Shewry, P. R. Improving the protein content and composition of cereal grain. *J. Cereal Sci.* **2007**, *46*, 239−250.
(11) Garvin, D. F.; Welch, R. M.; Finley, J. W. Historical shifts in the seed mineral micronutrient concentration of US hard red winter wheat germplasm. *J. Sci. Food Agric.* **2006**, *86*, 2213−2220.
(12) Oury, F.-X.; Leenhardt, F.; Rémésy, C.; Chanliaud, E.; Duperrier, B.; Balfourier, F.; Charmet, G. Genetic variability and stability of grain magnesium, zinc and iron concentrations in bread wheat. *Eur. J. Agron.* **2006**, *25*, 177−185.
(13) Murphy, K. M.; Reeves, P. G.; Jones, S. S. Relationship between yield and mineral nutrient concentrations in historical and modern spring wheat cultivars. *Euphytica* **2008**, *163*, 381−390.
(14) Fan, M.-S.; Zhao, F.-J.; Fairweather-Tait, S. J.; Poulton, P. R.; Dunham, S. J.; McGrath, S. P. Evidence of decreasing mineral density in wheat grain over the last 160 years. *J. Trace Elem. Med. Biol.* **2008**, *22*, 315−324.
(15) Poutanen, K.; Shepherd, R.; Shewry, P. R.; Delcour, J. A.; Björck, I.; van der Kamp, J.-W. Beyond whole grain: the European HEALTHGRAIN project aims at healthier cereal food. *Cereal Foods World* **2008**, *53*, 32−35.
(16) Ward, J. L.; Poutanen, K.; Gebruers, K.; Piironen, V.; Lampi, A.-M.; Nyström, L.; Andersson, A. A. M.; Åman, P.; Boros, D.; Rakszegi, M.; Bedő, Z.; Shewry, P. R. The HEALTHGRAIN cereal diversity screen: concept, results and prospects. *J. Agric. Food Chem.* **2008**, *56*, 9699−9709.
(17) Shewry, P. R.; Piironen, V.; Lampi, A.-M.; Edelmann, M.; Kariluoto, S.; Nurmi, T.; Fernandez-Orozco, R.; Ravel, C.; Charmet, G.; Andersson, A. A. M.; Åman, P.; Boros, D.; Gebruers, K.; Dornez, E.; Courtin, C. M.; Delcour, J. A.; Rakszegi, M.; Bedő, Z.; Ward, J. L. The HEALTHGRAIN wheat diversity screen: effects of genotype and environment on phytochemicals and dietary fiber components. *J. Agric. Food Chem.* **2008**, *56*, 9758−9766.
(18) Nurmi, T.; Nyström, L.; Edelmann, M.; Lampi, A.-M.; Piironen, V. Phytosterols in wheat genotypes in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2008**, *56*, 9710−9715.
(19) Lampi, A.-M.; Nurmi, T.; Ollilainen, V.; Piironen, V. Tocopherols and tocotrienols in wheat genotypes in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2008**, *56*, 9716−9721.
(20) Andersson, A. A. M.; Kamal-Eldin, A.; Frás, A.; Boros, D.; Åman, P. Alkylresorcinols in wheat varieties in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2008**, *56*, 9722−9725.
(21) Piironen, V.; Edelmann, M.; Kariluoto, S.; Bedő, Z. Folate in wheat genotypes in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2008**, *56*, 9726−9731.
(22) Li, L.; Shewry, P. R.; Ward, J. Phenolic acids in wheat varieties in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2008**, *56*, 9732−9739.

(23) Gebruers, K.; Dornez, E.; Boros, D.; Frás, A.; Dynkowska, W.; Bedő, Z.; Rakszegi, M.; Delcour, J. A.; Courtin, C. M. Variation in the content of dietary fiber and components thereof in wheats in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2008**, *56*, 9740−9749.

(24) Egli, D. B. *Seed Biology and the Yield of Grain Crops*; CAB International: Wallingford, Oxfordshire, U.K., 1998.

(25) Zhao, F. J.; Su, Y. H.; Dunham, S. J.; Rakszegi, M.; Bedő, Z.; McGrath, S. P.; Shewry, P. R. Variation in mineral micronutrient concentrations in grain of wheat lines of diverse origin. *J. Cereal Sci.* **2009**, *49*, 290−295.

(26) McDonald, G. K.; Genc, Y.; Graham, R. D. A simple method to evaluate genetic variation in grain zinc concentration by correcting for differences in grain yield. *Plant Soil* **2008**, *306*, 49−55.

(27) Morgounov, A.; Gomez-Becerra, H. F.; Abugalieva, A.; Dzhunusova, M.; Yessimbekova, M.; Muminjanov, H.; Zelenskiy, Y.; Ozturk, L.; Cakmak, I. Iron and zinc grain density in common wheat grown in Central Asia. *Euphytica* **2007**, *155*, 193−203.

(28) Andersson, A. A. M.; Kamal-Eldin, A.; Åman, P. Effects of environment and variety onalkylresorcinols in wheat in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2010**, *58*, 9299−9305.

(29) Lampi, A.-M.; Nurmi, T.; Piironen, V. Effects of the environment and genotype on tocopherols and tocotrienols in wheat in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2010**, *58*, 9306−9313.

(30) Nurmi, T.; Lampi, A.-M.; Nyström, Piironen, V. Effects of environment and genotype on phytosterols in wheat in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2010**, *58*, 9314−9323.

(31) Kariluoto, S.; Edelmann, M.; Piironen, V. Effects of environment and genotype on folate contents in wheat in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2010**, *58*, 9324−9331.

(32) Fernandez-Orozco, R.; Li, L.; Harflett, C.; Shewry, P. R.; Ward, J. L. Effects of environment and genotype on phenolic acids in wheat in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2010**, *58*, 9341−9352.

(33) Gebruers, K.; Dornez, E.; Bedö, Z.; Rakszegi, M.; Frás, A.; Boros, D; Courtin, C. M.; Delcour, J. A. Environment and genotype effects on the content of dietary fiber and its components in wheat in the HEALTHGRAIN diversity screen. *J. Agric. Food Chem.* **2010**, *58*, 9353−9361.

(34) Graham, R.; Senadhira, D.; Beebe, S.; Iglesias, C.; Monasterio, I. Breeding for micronutrient density in edible portions of staple food crops: conventional approaches. *Field Crops Res.* **1999**, *60*, 57−80.

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