

NOTE

Determination of Protein and 11 Elements in Six Milling Fractions of Two Wheat Varieties

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ABSTRACT

Cereal Chem. 64(4):285-287

The concentrations of Br, Ca, Cl, Fe, K, Mg, Mn, Na, Rb, Se, and Zn were measured in a hard red winter (Scout 66) and a soft red winter (Coker 762) wheat cultivar using neutron activation analysis. The soft wheat had a higher concentration of Br, Cl, Fe, K, Mn, and Zn and a lower concentration of Ca. Depending on the particle size, the ground whole wheat was separated into six milling fractions labeled A (coarse) through F

(fine). The highest concentrations of Ca, K, Mn, Na, Rb, Se, and Zn were found in fractions A and B. Except for Br and Cl, both cultivars showed positive correlations between the elements measured in the milling fractions. The *in vitro* protein digestibility for whole wheat was higher compared to that of fractions A and B, and lower compared to that of fractions C through F.

Cereals, particularly wheat, are a major source of energy, minerals, and protein in human diet. Pomeranz (1971) and Lorenz et al (1980) reported that upon milling the wheat, minerals are distributed unequally among the various milling fractions, and as a result, some minerals in wheat flour are reduced by milling. Pomeranz (1971) reported that 50% of the Ca is in the endosperm and 25% in the aleurone layers; less than 10% of the Mg and Zn are in the endosperm, with 70 and 50%, respectively, in the aleurone layers. The National Academy of Sciences (NAS/NRC 1974) proposed a fortification policy for cereal-grain products that included zinc and magnesium. The academy recommended further studies related to toxicity and deficiency before considering Cu, Cr, Se, and Mn for fortification. Lorenz et al (1980) reported that the low recovery of iron and calcium as a result of milling wheat into flour may require the addition of these elements as an enrichment in many countries.

El-Gindy et al (1957) studied the effect of agronomic growing conditions on mineral content of wheat. They reported correlation between fertilizer treatment and total ash of three hard red winter wheats grown in 13 locations utilizing seven different fertilizers. They also concluded that concentration of minerals depends on the varieties tested. Correlation between minerals in hard red winter wheat has been shown by Dikeman et al (1982). Numerous investigators including Cirilli (1971), Garcia et al (1972a,b), O'Dell et al (1972), Peterson et al (1983), Schweizer et al (1984), and Iskander and Morad (1986a,b) have reported mineral contents of various flour fractions and the effect of extraction rate. They found wheat bran to contain a higher percentage of minerals compared to whole wheat or wheat flour. Iskander and Morad (1986b) studied mineral distribution, protein content, and *in vitro* protein digestibility in the whole wheat and six milling fractions of four hard red winter wheat varieties. They concluded that the concentrations of Ca, Fe, Mg, Mn, and Zn were highest in the material retained by sieve no. 30 while the lowest concentration was in the material that passed through sieve no. 150.

Kent and Evers (1969) found that the outer 10–11% of the starchy endosperm accounts for 25% of the total endosperm protein in samples of hard red winter wheat. Jones et al (1959) stated that protein content of the coarse fractions was usually higher than that of the initial flour or that of fine fractions when air classification was used to fractionate wheat flour based on particle size.

The purposes of this study were to investigate mineral content using neutron activation analysis, protein content, and *in vitro* protein digestibility of a hard red winter wheat variety versus a soft red winter wheat variety grown in similar environmental and agronomic conditions. Distribution of minerals and protein in six milling fractions derived from the two varieties was also investigated.

MATERIALS AND METHODS

Materials

One hard red winter wheat (Scout 66) and a soft red winter wheat (Coker 762) variety grown in 1982 at Chillicothe, TX, were used. Environmental and agronomic conditions under which the two varieties were grown were similar. For each variety, 100 g was tempered to 15% moisture using distilled water for 18 hr, then milled using a Quadrumat Junior mill. A stack of five U.S. Standard testing sieves (nos. 30, 40, 70, 100, and 150) with three rubber bouncers inserted into each sieve was placed on a Ro-Tap sifter. The stack was agitated for exactly 3 min. The first five fractions—A, B, C, D, and E—were the “over” on each sieve and were collected, weighed, and stored in tightly closed containers. The sixth fraction, F, was the material collected “under” sieve 150 and was treated similarly.

Protein Content

Samples (whole wheat or milling fractions) were analyzed for nitrogen using the modified micro-Kjeldahl procedure (Technicon 1977). Protein was calculated as $N \times 5.7$.

In Vitro Protein Digestibility

In vitro protein digestibility was evaluated via pronase hydrolysis using the method of Hahn et al (1982).

Neutron Activation Analysis

A 300–450 mg sample was used to measure the elements of interest. Pre-irradiation sample manipulation, irradiation scheme, and post-irradiation counting procedures were the same as those used for other wheat cultivars in a study by Iskander and Morad (1986b).

Statistics

Data were statistically analyzed using one-way analysis of variance and Duncan's new multiple range test (Steel and Torrie 1960).

RESULTS AND DISCUSSION

The weight distributions of the milling fractions for the two

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wheat varieties are presented in Table I. For the soft wheat, 25.3% was separated in fraction A compared to 9.06% for the hard wheat. On the other hand, fraction F constituted 24.8% for the hard wheat compared to 7.24% for the soft. The weight of fractions B and D for both varieties did not vary significantly.

The concentrations of 11 elements in the whole wheat and the six milling fractions are reported in Table II. The soft wheat variety had higher concentrations of Br, Cl, Fe, K, Mn, and Zn and a lower concentration of Ca compared to the hard wheat variety. The difference in Mg, Na, Rb, and Se between the two varieties was not significant. Using dry weight basis, Dikeman et al (1982) reported the following element concentrations for Scout 66 ($\mu\text{g/g}$): Ca, 515; Fe, 39.2; K, 4,440; Mg, 1,500; Mn, 47.5; and Zn, 23.6. Considering the moisture content, our results on Fe, Mg, and Zn vary from this study by approximately 10%. The large difference between K concentration reported by Dikeman (1982) and data reported in this study could be explained by differences in fertilization or environmental conditions.

Another 13 elements were measured in the two wheat varieties but were found to be lower than the detection limit under the experimental conditions. The upper concentrations ($\mu\text{g/g}$) between the two varieties were as follows: As <0.1, Ce <0.1, Co <0.04, Cs <0.009, Eu <0.002, Hf <0.01, Hg <0.05, La <0.04, Sb <0.05, Sc <0.001, Sr <5.00, Th <0.007, and U <0.01.

The concentrations of Br, Ca, Cl, Fe, K, Mg, Mn, Na, Rb, Se, and Zn in the six milling fractions are also shown in Table II. For soft wheat, Br shows equal distribution in all the fractions, whereas in the hard variety fraction F contained the highest Br concentration. Also, Cl tended to concentrate in the fine fractions (D, E, and F) in the hard wheat variety and in E and F in the soft wheat variety. The distribution of Ca, Fe, K, Mg, and Mn, Na, Rb, Se, and Zn follows approximately the same pattern, where the highest concentration of the element was found in fraction A followed by fraction B.

The correlation coefficient (r) between the elements in the milling fractions for each variety was computed. For the hard wheat variety, a positive correlation ($r > 0.90$) was observed between Ca, Fe, K, Mg, Mn, Na, Rb, Se, and Zn. Both Br and Cl were negatively correlated with the nine elements ($r < -0.46$ for Br, and $r < -0.76$ for Cl). For the soft wheat variety, a less positive correlation ($r > 0.78$) was observed between the aforementioned group of nine elements. Br and Cl were negatively correlated with the other elements in the soft variety ($r < -0.33$ for Br, and $r < -0.39$ for Cl).

The concentration of protein in the whole wheat and the six milling fractions (Table III) is in decreasing order A, B, F, whole wheat, C, D, and E. This order is the same for both soft and hard varieties, which indicates that protein tends to concentrate in the coarse fractions A and B. This could be explained by the presence

of the aleurone layers that separated mainly in the first two fractions. However, the in vitro protein digestibility of the coarse fractions A and B in both varieties was lower compared to other fractions and whole wheat, probably because of the presence of a higher level of fiber and pentosans in the larger particle size fractions. In general, the hard red winter wheat variety examined in this study contained higher protein content but of lower in vitro digestibility compared to the soft wheat variety.

The protein content was positively correlated to Ca, Fe, K, Mg, Mn, Na, Rb, Se, and Zn ($r > 0.90$ for hard wheat, and $r > 0.87$ for soft wheat) and negatively correlated to Br ($r = -0.37$ for hard wheat, and $r = -0.48$ for soft wheat) and Cl ($r = -0.76$ for hard wheat, and $r = -0.58$ for soft wheat). The correlation between in vitro protein digestibility and the concentrations of the nine elements was negative, with $r < -0.95$ for hard wheat and $r < -0.91$ for soft wheat. For Br and Cl, the respective values of r were 0.49 and 0.87 for hard wheat and 0.55 and 0.54 for soft wheat.

CONCLUSION

The concentrations of Mg, Na, Rb, and Se in ground whole wheat of the two varieties did not vary significantly. The concentrations of Br, Cl, Fe, K, Mn, and Zn were higher in the soft wheat; however, the soft wheat variety was lower in Ca. In both varieties, the highest concentrations of Ca, Fe, K, Mg, Mn, Rb, Se, and Zn were found in the coarse fractions compared to the fine fractions. Because different milling fractions are used in various baking industries and because the mineral content is often reduced upon milling wheat into flour, the knowledge of element concentration should help identify the elements that could potentially be used for enrichment. However, human safety must be considered before any recommendation is made in regard to adding any individual element to flour.

This study focused only on two wheat varieties. However, two of us performed similar studies on four hard red winter wheat varieties (Iskander and Morad 1986a,b). Further studies to include a greater number of wheat classes grown under different environmental conditions are recommended.

TABLE I
Weight of Six Milling Fractions as a Percent of Whole Wheat for a Hard Red Winter (Scout 66) and a Soft Red Winter (Coker 762) Wheat

Fraction	A	B	C	D	E	F
Sieve	+30 ^a	+40	+70	+100	+150	-150 ^b
Hard wheat (Scout 66)	9.06	4.54	22.6	15.7	23.6	24.5
Soft wheat (Coker 762)	25.3	4.86	29.4	14.6	18.6	7.24

^a+ = Materials retained on and collected over the sieve.

^b- = Materials passed through and collected under the sieve.

TABLE II
Concentration of 11 Elements in Whole Wheat and in Six Milling Fractions of Hard Red Winter (Scout 66) and Soft Red Winter (Coker 762) Wheat Varieties

Sample ^b	Br	Ca	Cl	Fe	K	Mg	Mn	Na	Rb	Se	Zn
Hard Wheat (Scout 66)											
Whole wheat	4.95	562	494	37.3	3,040	1,260	31.0	14.7	1.98	0.58	18.6
Fraction A	10.7	1,420	454	132	11,600	5,280	144	42.0	4.74	0.79	56.5
Fraction B	10.1	1,170	417	135	9,000	3,550	130	34.9	5.12	0.69	67.9
Fraction C	11.4	282	522	17.6	1,890	726	17.7	9.35	1.31	0.42	12.6
Fraction D	12.0	274	742	19.3	1,660	382	9.42	8.92	1.40	0.42	9.58
Fraction E	13.2	327	698	25.1	1,500	439	7.42	15.2	1.20	0.47	8.54
Fraction F	16.8	287	701	32.0	1,460	341	5.53	15.1	1.15	0.49	10.8
Soft Wheat (Coker 762)											
Whole wheat	7.05	397	527	42.5	4,060	1,340	38.7	15.8	1.94	0.50	21.3
Fraction A	11.3	904	672	126	9,150	4,550	93.3	36.7	3.78	0.44	55.6
Fraction B	10.1	640	690	112	5,920	2,290	79.2	25.8	4.91	0.46	77.7
Fraction C	11.2	224	732	32.9	2,110	478	13.6	10.2	1.37	0.29	14.2
Fraction D	11.5	160	731	20.2	1,720	432	6.05	8.39	0.75	0.28	6.59
Fraction E	11.5	187	1,080	35.2	1,700	357	6.77	15.5	<0.7	0.28	7.54
Fraction F	11.5	245	990	26.2	1,530	362	6.51	13.5	<0.6	0.34	8.53

^aConcentration in $\mu\text{g/g}$. All values reported on 15% moisture basis. $n = 2$.

^bFractions are defined in Table I.

TABLE III
Protein Content and In Vitro Protein Digestibility
of Hard Red Winter (Scout 66) and Soft Red Winter (Coker 762)
Wheat Varieties and Respective Milling Fractions^a

Sample ^b	Hard Wheat (Scout 66)		Soft Wheat (Coker 762)	
	Protein (%)	In Vitro Protein Digestibility (%)	Protein (%)	In Vitro Protein Digestibility (%)
Whole wheat	13.4 c	62.3 bc	12.8 c	76.6 b
Fraction A	17.7 a	58.3 c	16.8 a	62.6 cd
Fraction B	17.6 a	58.6 c	15.2 b	67.8 c
Fraction C	12.6 d	67.1 ab	12.4 cd	87.2 a
Fraction D	12.4 d	70.2 a	12.3 d	85.4 a
Fraction E	13.3 c	67.1 ab	12.0 d	86.0 a
Fraction F	14.5 b	66.4 ab	12.9 c	82.7 ab

^aNitrogen content on 15% moisture basis. Conversion factor is 5.7. $n = 2$. Means with the same letters in each column are not significantly different ($P < 0.05$).

^bFractions are defined in Table I.

ACKNOWLEDGMENTS

This work was presented at the AACC 70th Annual Meeting in Orlando, FL, September, 1985. Technical assistance from Michael G. Krause and Bonnie J. Malek is deeply appreciated.

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[Received November 20, 1985. Revision received January 30, 1987. Accepted March 10, 1987]