

Flavonoids and Antioxidative Activities in Buckwheat

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Samples of buckwheat from four cultivars grown at three locations in western Canada for 4 years were used to study the effect of cultivar, location, and growing season on the flavonoid content and antioxidative activities of the seed. Buckwheat contained an average of 387 and 1314 mg/100 g of flavonoid and 47 and 77 mg/100 g of rutin in the seed and hull, respectively. Location was the main source of variation for flavonoid and rutin contents of the seed, while growing season had significant influence on the flavonoid content of the hulls. Variation in antioxidative activities was mainly due to a cultivar \times environment effect. Antioxidative activities expressed as AOX (Δ log A470/min), AA (% inhibition relative to control), and ORR (oxidation rate ratio) ranged from 0.42, 114, and 0.16 to 1.63, 48, and 0.59, respectively. Flavonoid content in buckwheat was strongly correlated with rutin content and weakly associated with antioxidative activities, while rutin content was not related to antioxidative activities.

Keywords: *Buckwheat; flavonoid; rutin; antioxidant; cultivar effects; seasonal variations; Fagopyrum esculentum*

INTRODUCTION

Buckwheat (*Fagopyrum esculentum* Moench) is an important alternative crop in western Canada with an estimated farmgate value of about \$5.5 million. Much of the grain is exported to Japan where it is used to manufacture soba noodles and other consumer products (Mazza, 1993). For Canadian buckwheat producers to remain competitive in world markets, they must continue to improve the quality of their product. As part of this effort, development of cultivars with improved nutritional and storage stability is imperative. Characteristics of importance for storage stability include the concentration of endogenous antioxidants such as flavonoids and phenolics. Cultivars differ in these characteristics, which are also influenced by environmental factors (Oomah et al., 1995; Kitabayashi et al., 1995).

Buckwheat is considered to be a major dietary source of rutin. Hence interest in buckwheat is increasing because of the possible beneficial health effects of rutin. Activities demonstrated by rutin include antiinflammatory, antimutagenic, antitumoral, anticarcinogenic, smooth muscle relaxation, and estrogen receptor binding (Pisha and Pezzuto, 1994). Rutin is also known to reduce fragility of blood vessels associated with some hemorrhagic diseases or hypertension in humans (Griffith et al., 1944; Matsubara et al., 1985; Iwata et al., 1990; Yildzogle-Ari et al., 1991). Under conditions of low dietary fat intake, phenolic flavonoids such as rutin and quercetin have been reported to considerably suppress colon tumor incidence (Deschner, 1992).

Varietal differences and heritability of rutin content in common buckwheat have been attributed to geographic origin of the seed as well as environmental conditions (Kitabayashi et al., 1995). Kitabayashi et al. (1995) reported that rutin content in buckwheat seed

ranges from 12.6 (for a Russian cultivar) to 35.9 mg/100 g dry weight for a Nepalese strain.

Improvement of rutin content in buckwheat is an important breeding objective in Japan, and high rutin content lines of common buckwheat have been developed and are being tested for adaptability (Ohsawa and Tsutsumi, 1995). Several factors affect the accumulation of flavonoids in buckwheat. Ohara et al. (1989) suggested that the varying amount of solar radiation accounted for the variation in rutin content. Cropping season such as day length conditions increased the accumulation of rutin in buckwheat (Ohsawa and Tsutsumi, 1995). In addition, late flowering cultivars under Japanese conditions had higher rutin content than early flowering cultivars.

This study was conducted to investigate the variation of flavonoid and rutin content due to location in different buckwheat cultivars grown in western Canada. A second objective was to examine the antioxidative response of various buckwheat cultivars to assess the prospects of improving the storability by agronomic and/or genetic methods.

MATERIALS AND METHODS

Samples of four buckwheat cultivars (AC Manisoba, BS85601, CM-15, and Manor) were obtained from cooperative tests conducted at three locations (Brooks, Alberta; Morden, Manitoba; and Scott, Saskatchewan) in western Canada during the 1990–1993 growing seasons. The tests were standardized according to procedures established by the Western Expert Committee on Grain (Anonymous, 1991). Hulls were obtained by dehulling buckwheat seed between a stationary and a rotating emery stone as described by Mazza and Campbell (1985).

Whole buckwheat was ground in a Wiley mill to pass a 1 mm screen. The spectrophotometric assay for the quantitative determination of flavonoid content was carried out, essentially, as described by Hairi et al. (1991). Briefly, ground buckwheat (1 g) was extracted with 80% methanol (25 mL) on a wrist shaker for 120 min at 71 °C. The extract was filtered (Whatman No. 5) on a Büchner funnel. The filtrate (1 mL) was diluted twice with distilled water, and absorption was

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measured at 404 nm (DU-50, Beckman, Beckman Instruments, Inc., Irvine, CA) after the addition of 100 μ L of 1% (2-aminoethyl)diphenylborate solution. Extract absorption was compared to that of a standard rutin (Sigma Chemical Co., St. Louis, MO) curve.

Rutin content was determined by high-performance liquid chromatography (HPLC) following a modification of the procedure described by Terada and Miyabe (1993). For a typical assay, a 5 mL aliquot of the filtered extract (obtained for the determination of flavonoid described above) was dried under vacuum at 30 °C, dissolved in 1 mL of methanol–water–oxalic acid (13/36/1 v/v/v), and filtered through a 0.45 μ m Millipore filter prior to HPLC analysis. Samples (100 μ L) were analyzed by using an HPLC system consisting of two LKB pumps Model 2150, a solvent conditioner (LKB, Model 2156), a column temperature controller (Waters-TCM model, Millipore Corp., Milford, MA), and an LKB variable wavelength UV monitor (Model 2141, Pharmacia, Bromma, Sweden) interfaced through a PE Nelson 900 series with a personal computer. Separations were performed on a 3 μ m (100 \times 4.6 mm i.d.) CapCell Pak C₁₈-SG120 column (Dychrom, Santa Clara, CA). Chromatographic runs were carried out using a gradient of solvents A [MeOH/H₂O/CH₃COOH 13/36/1 (v/v/v)] and B [MeOH/H₂O/CH₃COOH 73/25/2 (v/v/v)] at 40 °C. The following gradient was used: linear from 10% B to 50% B during 20 min followed by isocratic 50% B over 5 min and a return to 10% B in 5 min. The flow rate was 0.5 mL/min. Typically, a 10 min equilibrium period was used between samples. Detection of rutin was measured at 350 nm. Quantitation was based on an external standard method where the calibration curve ranged from 1 to 250 mg/mL of rutin (Sigma Chemical Co., St. Louis, MO) using Model 2600 Chromatography Software, revision 3.1 (Nelson Analytical, Inc., Cupertino, CA).

Heat-induced oxidation of an aqueous emulsion system of β -carotene and linoleic acid was used as the antioxidant activity test model (Pratt, 1992). One milliliter of β -carotene (0.2 mg/mL dissolved in chloroform; Sigma Chemical Co., St. Louis, MO) was added to Erlenmeyer flasks containing linoleic acid (0.02 mL) and Tween 20 (0.2 mL). Each mixture was then dosed with 0.2 mL of the corresponding filtered flavonoid extract or butylated hydroxytoluene (15 mg/L). Samples without dosed compounds were used as standard. Each sample was concentrated using N₂ in the dark. Fifty milliliters of distilled water, saturated with air (0.5 L/min, 15 min), was added to each mixture, and the resulting mixture was shaken. Their absorbances were measured on a spectrophotometer (Beckman DU-50) at 470 nm. The samples were then subjected to thermal autoxidation by keeping them in a constant temperature water bath at 50 °C for 2 h. The rate of bleaching of β -carotene was monitored by taking the absorbance at 10 min intervals. Antioxidative activity was calculated by three different methods. For the first method, the log of the absorbance was plotted against time, as a kinetic curve, and the slope was expressed as the AOX value. The second method of calculation based on first order kinetics was conducted as described by Al-Saikhan et al. (1995) using the following equations:

$$\ln(a/b) \times 1/5 = \text{sample degradation rate}$$

where \ln = natural log; a = initial absorbance at 470 nm and at time 0; b = absorbance at 470 nm and at 10, 20, and 30 min; 5 = time (min). Antioxidant activity (AA) was also calculated as % inhibition relative to the control using the following relationship:

$$\text{AA} = \frac{\text{degradation rate of control} - \text{degradation rate of sample}}{\text{degradation rate of control}} \times 100$$

where BHT was the control. The AA values for different times were averaged to give one AA value for the sample.

Table 1. Total Flavonoid and Rutin Contents (mg/100 g) of Buckwheat Seed and Hulls

cultivar	flavonoids		rutin		hull (%)
	seed	hull	seed	hull	
AC Manisoba	372.8 ^b	1302.8 ^b	44.7 ^b	76.1 ^c	25.6 \pm 2.7
BS85601	396.5 ^a	1277.7 ^b	46.4 ^b	85.3 ^b	25.7 \pm 3.2
CM-15	371.5 ^b	1212.8 ^c	44.2 ^b	50.5 ^d	34.0 \pm 2.1
Manor	407.5 ^a	1463.7 ^a	51.1 ^a	97.4 ^a	26.4 \pm 3.0

^{a-d} Means in a column followed by the same letter are not significantly different by Duncan's multiple range test at the 5% level.

The third method of expression based on the oxidation rate ratio, ORR, was calculated by the method of Marinova et al. (1994) using the equation

$$\text{ORR} = \text{Bs/Bo}$$

where Bs is the rate of bleaching of β -carotene in the presence of the sample, and Bo is the rate of bleaching in the absence of the sample (blank).

Protein content ($N \times 5.53$) was determined on dehulled, ground, and oven dried buckwheat by the Kjeldahl method with a Tecator digester and a Kjeltac (System 1002) distillation unit (Tecator AB, Höganäs, Sweden).

Analysis of variance by the general linear models (GLM) procedure, means comparison by Duncan's test, Pearson correlation, and variance components using PROC VARCOMP procedure were performed according to SAS methods (SAS, 1990). All effects were considered random for the variance component analysis, and calculations were based on type I sum of squares method.

RESULTS AND DISCUSSION

Total flavonoid contents of buckwheat grown at 3 locations in 1991 differed significantly among cultivars ($P = 0.05$) (Table 1). However, cultivars AC Manisoba and CM-15 had similar (372 mg/100 g), but significantly lower, flavonoid contents than those of BS85601 and/or Manor. Flavonoids were highly concentrated in the hulls which contained over 3 times the level present in the seed. Thus, the mean flavonoid contents of buckwheat seed and hulls were 387 and 1314 mg/100 g, respectively. Interestingly, a high level of hulls in cv. CM-15 (34%) corresponded to the lowest concentrations of flavonoid and rutin in both the seed and hulls (Table 1). Cultivar Manor which had the highest flavonoid and rutin content in the seed also had the maximum concentration of these compounds in the hulls. Rutin content of buckwheat hulls differed significantly among cultivars although its concentration was similar in the seed except for cv. Manor. Rutin content of hulls was about 1 1/2 times that in the seed with a mean of 47 and 77 mg/100 g for seed and hulls, respectively.

To further elucidate the variability in buckwheat flavonoid and rutin contents, environment (location) and seasonal (year) effects were studied in combination with cultivars. The results of analysis of variance for flavonoid and rutin contents of buckwheat grown at 3 locations for 4 years (Table 2) were parallel and showed that flavonoid and rutin contents were dependent on cultivar, location, and year and their interactions. Environmental effect, i.e., location, and year \times location interaction had a much larger relative contribution to the variation in flavonoid and rutin contents than cultivar or seasonal effects. Location and year \times location interaction explained about 50% and 20% of the variation, respectively, in flavonoid and rutin contents in buckwheat (Table 2). Cultivar and its interactions, cultivar \times location, cultivar \times year, and cultivar \times year,

Table 2. Analysis of Variance for Total Flavonoids (Buckwheat Seed and Buckwheat Hulls) and Rutin of Buckwheat Seed Grown at Three Locations in 4 Years

source	df	mean squares ^a		
		total flavonoid		rutin
		whole seed	hulls	
cultivar (C)	3	1.99 (8.5)	29.93 (11.8)	508.88 (12.0)
location (L)	2	14.87 (52.9)	61.03 (16.9)	2020.14 (49.8)
year (Y)	3	2.30 (4.9)	69.59 (39.0)	390.43 (2.7)
C × Y	8	0.38 (1.2)	6.73 (9.1)	26.83 ^c (0)
C × L	6	0.10 ^c (0)	1.39 ^b (0)	35.04 ^c (0.7)
Y × L	4	1.57 (18.6)	10.66 (13.2)	318.11 (20.4)
C × Y × L	9	0.30 (9.7)	2.39 (7.1)	69.55 (9.4)
error	36	0.04 (4.2)	0.35 (2.9)	10.70 (5.0)
CV %		5.00	4.56	7.08

^a All mean squares are significant at 0.0001 probability levels, except *b* and *c* at 0.005 and 0.05, respectively. Values in parentheses are percent variance components.

× location had very small effects on flavonoid and rutin contents. Cultivar was also reported (McGregor et al., 1952) to have little or no effect on the rutin content of 17 strains of buckwheat tested for a 3 year period. Recently, Kitabayashi et al. (1995) observed varietal and year effects in 27 cultivars and strains of buckwheat grown at one location for 2 years. Differences in rutin content of buckwheat seed were found to be highly significant ($P = 0.01$) among locations (between areas) and cultivars, while the seasonal effect was only significant at a 1% level (Kitabayashi et al., 1995). Our results, indicating that variation of rutin content among buckwheat cultivars is mainly due to environmental factors, are consistent with those of Kitabayashi et al. (1995).

Variability in flavonoid content of buckwheat hulls was due to the main effects, year, location, and cultivar, which accounted for 39%, 17%, and 12% of the total variation, respectively (Table 2). It is obvious that differences in years were more remarkable than cultivar and location for hull flavonoid as compared to location for both flavonoid and rutin contents of the seed. The cultivar × year interaction was a significant source of variation for hull flavonoid but not for flavonoid and rutin contents of the seed. The variation components for cultivar × year for flavonoid contents of hulls and seed were 9.1% and 1.2%, respectively (Table 2). The cultivar × location interaction does not play a significant role in the variability of flavonoid and rutin contents since the variance component was nil.

Location was highly significant for rutin content (Table 2), so cultivar differences were examined sep-

arately for the three locations (Table 3). Generally, buckwheat grown in Scott, Saskatchewan, had higher rutin concentration in both the seed and the hulls than those samples grown in Brooks and Morden. Rutin content of buckwheat grown in Scott was 68–91% and 25–76% higher than that of buckwheat grown in Morden and Brooks, respectively. Hulls of buckwheat grown in Scott had 2.3–2.7 and 1.2–1.6 times higher rutin concentration than those of samples grown in Morden and Brooks, respectively. Cultivars AC Manisoba and BS85601 had similar levels of rutin content at each location. The ratio of rutin content in hulls and seed for these two cultivars increased from 1.4 to 1.6 to 1.9 for Morden, Brooks, and Scott, respectively. Manor had the highest level of rutin content in the seed and hulls but was the least stable cultivar regarding rutin content. The exceptionally low rutin content of CM-15 hulls at Morden cannot be explained completely, since repeated extractions (4×) yielded the same values. The differences in rutin content at various locations are probably associated with soil types and conditions and temperature and moisture stress. Brooks, Scott, and Morden are located in the brown, dark brown, and black soil zones, respectively. Furthermore, it was observed that local factors also affected seed yield inversely to those of rutin content. For example, in 1991, the average yield of the four buckwheat cultivars grown in Brooks, Morden, and Scott were 2130 ± 499 , 1339 ± 145 , and 602 ± 25 kg/ha, respectively. Rutin as well as flavonoid contents correlated significantly ($r = -0.76$, $P = 0.05$ and $r = -0.89$, $P = 0.01$, respectively) with yield of buckwheat grown at Scott. This suggests that environmental stress which depresses yield could be responsible for the increased accumulation of rutin in buckwheat seed. Similar effects on rutin content due to soil types and conditions were reported in buckwheat seeds (Kitabayashi et al., 1995) and leaves (Hagels et al., 1995). These results agree with those of Patil et al. (1995) who observed similar changes in quercetin concentration in onion due to location and soil type.

Antioxidative activity, based on the inhibition of linoleic acid autoxidation, and expressed as AOX (Δ log A470/min), AA (% inhibition relative to control), and ORR (ratio) values showed similar trends. In the analysis of variance, there were differences among cultivars and years, and cultivar × location and year × location interactions were also significant (Table 4). Most of the variance was associated with cultivar × location and year × location interactions for AOX, AA, and ORR. AOX and AA values were also dependent on years while AA and ORR values showed significant differences due to location. From the variance components, year, year × location, and cultivar × location interactions accounted for 64%, 59%, and 65% of the total variation in AOX, AA, and ORR values, respectively, thereby confirming the biological significance of these sources of variation in the experiment. Because

Table 3. Rutin Content (mg/100 g) of Buckwheat Hulls and Buckwheat Grown at Three Locations in 1991

cultivar/location	seed			hulls		
	Morden	Brooks	Scott	Morden	Brooks	Scott
AC Manisoba	32.0 ^c	45.5 ^{a,b}	56.7 ^b	46.3 ^a	73.9 ^b	108.8 ^{a,b}
BS85601	31.5 ^c	47.5 ^a	60.2 ^b	44.3 ^a	76.6 ^b	121.5 ^a
CM-15	35.5 ^b	37.8 ^b	59.7 ^b	14.7 ^b	56.8 ^b	89.9 ^b
Manor	38.3 ^a	41.7 ^{a,b}	73.3 ^a	52.3 ^a	101.7 ^a	126.9 ^a
mean of location	34.3 ^z	43.1 ^y	62.3 ^x	36.9 ^z	77.3 ^y	112.9 ^x

^{a–d,x–z} Means in a column for location or a row for seed and hulls, respectively, followed by the same letter are not significantly different by Duncan's multiple range test at the 5% level.

Table 4. Analysis of Variance for Antioxidative Activities of Buckwheat Grown at Three Locations in 4 Years

source	df	mean squares		
		AOX	AA	ORR ($\times 10^{-2}$)
cultivar (C)	3	0.35 ^c (8.3)	695.02 ^a (0)	7.32 ^a (8.3)
location (l)	2	0.18 (0)	823.88 ^a (0)	3.57 ^b (0)
year (y)	3	0.43 ^b (13.4)	662.78 ^a (12.8)	3.22 ^a (0)
C \times Y	6	0.04 (0)	114.96 (0)	0.64 (0)
C \times L	5	0.23 ^d (21.3)	856.79 ^a (29.6)	5.02 ^a (34.8)
Y \times L	4	0.24 ^d (28.2)	559.24 ^a (17.0)	5.13 ^a (30.3)
C \times Y \times L	1	0.11 (0)	582.02 ^b (32.1)	1.32 (15.1)
error	26	0.05 (28.8)	29.93 (8.5)	0.30 (11.5)

^{a-d} Mean squares significant at 0.0001, 0.001, 0.005, and 0.01, respectively.

of significant cultivar \times location interaction, antioxidative activities of the cultivars are presented (Table 5) rather than means across cultivars. Mean AOX, AA, and ORR values ranged from 0.42, 113.7, and 0.16, respectively, for Manor grown at Scott, to 1.63, 48.0, and 0.59, respectively, for BS85601 grown at Morden. The overall mean AOX value (1.18) was markedly different among cultivars but not among locations. The overall means for AA and ORR values (69.4 and 0.43, respectively) differed significantly among cultivars and within locations. When ORR, a measure of antioxidative strength (Marinova et al., 1994), is larger than 1, the oxidation proceeds faster in the presence of an inhibitor than in its absence. Thus, the lower the ORR value, the stronger the inhibitor. Buckwheat antioxidants, depending on cultivar and location of production, are inhibitors of a great strength because under the same conditions for BHT ORR = 0.19 ± 0.08 . The range of antioxidative activities of buckwheat cultivars is depicted in Figure 1.

Comparison of flavonoid and rutin with protein contents of buckwheat (data for protein not provided)

Table 5. Antioxidative Activities (AOX, AA, and ORR Values) of Buckwheat Grown at Three Locations for 4 years

cultivar/location	AOX ^f			AA ^g			ORR ^h		
	Morden	Brooks	Scott	Morden	Brooks	Scott	Morden	Brooks	Scott
AC Manisoba	1.29 ^{a,b}	1.10 ^{a,b}	1.47 ^a	70.7 ^a	76.1 ^{a,b}	66.3 ^{b,c}	0.41 ^b	0.39 ^{a,b}	0.47 ^{a,b}
BS85601	1.63 ^a	0.92 ^b	1.17 ^b	48.0 ^b	83.6 ^a	71.0 ^b	0.59 ^a	0.31 ^b	0.40 ^b
CM-15	1.19 ^b	1.38 ^a	1.29 ^{a,b}	70.6 ^a	65.1 ^c	59.0 ^c	0.45 ^b	0.47 ^a	0.55 ^a
Manor	1.13 ^b	1.00 ^b	0.42 ^c	63.6 ^a	71.8 ^{b,c}	113.7 ^a	0.44 ^b	0.39 ^{a,b}	0.16 ^c
mean of location	1.27 ^d	1.11 ^d	1.15 ^d	64.6 ^e	74.0 ^d	71.3 ^d	0.47 ^d	0.39 ^e	0.45 ^d

^{a-c} Means in a column followed by the same letter are not significantly different by Duncan's multiple range test at the 5% level. ^{d,e} Means in a row for each antioxidative activity followed by different letters are significantly different by Duncan's multiple range test at the 5% level. ^f AOX value = $\Delta \log A470/\text{min}$. ^g AA value = antioxidant activity expressed as % inhibition relative to control. ^h ORR value = oxidation rate ratio.

Table 6. Correlation Coefficients for Flavonoids, Rutin Content, and Antioxidative Activities of Buckwheat

	flavonoids for whole seeds	flavonoids for hulls	rutin content	antioxidative activities		
				AOX ^e	AA ^f	ORR ^g
protein	0.337		0.288			
flavonoids for whole seed		0.703 ^d	0.796 ^d	-0.436 ^c	0.447 ^b	-0.360 ^a
flavonoids for hulls			0.800 ^d	-0.444 ^b	0.379 ^a	-0.275
rutin content				-0.327	0.315	-0.262
AOX					-0.863 ^d	0.862 ^d
AA						-0.938 ^d

^a $P < 0.01$. ^b $P < 0.001$. ^c $P < 0.05$. ^d $P < 0.0001$ ($n = 52$ except for protein where $n = 30$). ^e AOX value = $\Delta \log A470/\text{min}$. ^f AA value = antioxidant activity expressed as % inhibition relative to control. ^g ORR value = oxidation rate ratio.

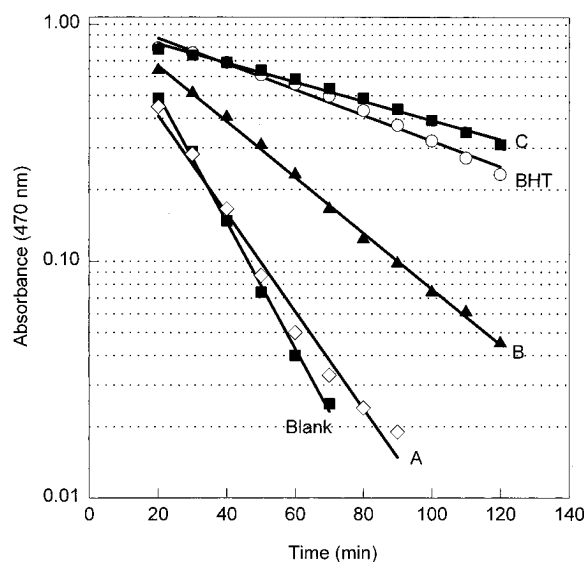


Figure 1. Antioxidative activities of (A) cultivar BS85601 grown at Morden in 1991, (B) Manor grown at Morden in 1992, and (C) Manor grown at Scott in 1990 compared to BHT and blank. The AOX values expressed as $\Delta \log A470/\text{min}$ for A, B, C were 2.21, 1.18, and 0.41, respectively. The AA values expressed as % inhibition relative to control for A, B, C were 20.6, 60.9, and 114.2, respectively. The ORR values, oxidation rate ratio, for A, B, C were 0.82, 0.46, and 0.15, respectively.

showed poor correlation. The Pearson correlation coefficients of flavonoid and rutin were 0.337 and 0.288, respectively (Table 6). The weak association between flavonoid, rutin, and protein content suggests that in buckwheat changes in flavonoid and rutin contents should have very little effect on protein content. Flavonoid contents of both whole seeds and hulls were highly correlated with rutin content but weakly associated with antioxidative activities. Nonsignificant correlation coefficients between rutin content and antioxidative activities suggest that they are totally independent of rutin concentrations in buckwheat. Nevertheless, cultivar Manor, grown at Scott and which had the highest rutin concentration in the seed and in the hulls (Table 3), also showed the highest antioxidative activities (Table 5). As expected, antioxidative activities

AOX, AA, and ORR were highly correlated with each other. When rutin content from three buckwheat hull samples (with 13.7, 36.6, and 42.7 mg/100 g sample) were compared with their antioxidative activities, high correlation coefficients ($r = 0.999$, $P < 0.05$) were obtained. This suggests that components other than flavonoids present in buckwheat seed probably contribute substantially to the antioxidative activities.

The data presented indicate that buckwheat cultivars differ in flavonoid and rutin contents and antioxidative activities. However, the narrow range of variations especially in rutin content of North American cultivars is to be expected due to its adaptation in northern latitudes. Since the interaction of cultivars with locations and environments was large, breeding of stable cultivars that interact less with the environments in which they grow should be emphasized to develop a cultivar with high levels of flavonoids. As flavonoids are found to be accumulating preferentially in the hulls, it may be used as a dietary source of rutin or it can be separated and incorporated as a source of dietary fiber rich in flavonoids.

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LITERATURE CITED

- Al-Saikhan, M. S.; Howard, L. R.; Miller, J. C., Jr. Antioxidant activity and total phenolics in different genotypes of potato (*Solanum tuberosum* L.). *J. Food Sci.* **1995**, *60*, 341–347.
- Anonymous. Requirements for registration of a buckwheat cultivar for production in Canada. In *The Prairie Registration Recommending Committee for Grain. Special Crops and Oilseed Crops. Minutes of the First Meeting*; Calgary, AB, Canada, 1991; pp 153–156.
- Deschner, E. E. Dietary quercetin and rutin: Inhibitors of experimental colonic neoplasia. In *Phenolic Compounds in Food and their Effects on Health II: Antioxidants and Cancer Prevention*; Huang, M-T., Ho, C-T., Lee, C. Y., Eds.; American Chemical Society: Washington, DC, 1992; pp 265–268.
- Griffith, J. Q.; Couch, J. F.; Lindauer, M. A. Effect of rutin on increased capillary fragility in man. *Proc. Soc. Exptl. Biol. Med.* **1944**, *55*, 228–229.
- Hagels, H.; Wagenbreth, D.; Schilcher, H. Phenolic compounds of buckwheat herb and influence of plant and agricultural factors (*Fagopyrum esculentum* Moench and *Fagopyrum tataricum* Gärtner). In *Current Advances in Buckwheat Research, Vol. II*; Matano, T., Ujihara, A., Eds.; Shinshu University Press: Matsumoto, Japan, 1995; pp 801–809.
- Hairi, B.; Sallé, G.; Andary, C. Involvement of flavonoids in the resistance of two poplar cultivars to mistletoe (*Viscum album* L.). *Protoplasma* **1991**, *162*, 20–26.
- Iwata, K.; Miwa, S.; Inayama, T.; Sasaki, H.; Soeda, K.; Sugahara, T. Effects of kangra buckwheat on spontaneously hypertensive rats. *J. Kagawa Nutr. Coll.* **1990**, *21*, 55–61.
- Kitabayashi, H.; Ujihara, A.; Hirose, T.; Minami, M. Varietal differences and heritability for rutin content in common buckwheat, *Fagopyrum esculentum* Moench. *Jpn. J. Breed.* **1995**, *45*, 75–79.
- Marinova, E. M.; Yanishlieva, N.; Kostova, I. N. Antioxidative action of the ethanolic extract and some hydroxycoumarins of *Fraxinus ornus* bark. *Food Chem.* **1994**, *51*, 125–132.
- Matsubara, Y.; Kumamoto, H.; Iizuka, Y.; Murakami, T.; Okamoto, K.; Miyake, H.; Yokoi, K. Structure and hypotensive effect of flavonoid glycosides in citrus unshiu peelings. *Agric. Biol. Chem.* **1985**, *49*, 909–914.
- Mazza, G.; Campbell, C. G. Influence of water activity and temperature on dehulling of buckwheat. *Cereal Chem.* **1985**, *62*, 31–34.
- Mazza, G. Buckwheat. In *Encyclopedia of Food Science, Food Technology and Nutrition, Vol. 1*; Macrae, R., Robinson, R. K., Sadler, M. J., Eds.; Academic Press: Toronto, Canada, 1993; pp 516–521.
- McGregor, W. G.; McKillican, M. E. Rutin content of varieties of buckwheat. *Sci. Agric.* **1952**, *32*, 48–51.
- Ohara, T.; Ohinata, H.; Muramatsu, N.; Matsubashi, T. Determination of rutin in buckwheat foods by high performance liquid chromatography. *Nippon Shokuhin Kogyo Gakkaishi* **1989**, *36*, 114–120.
- Ohsawa, R.; Tsutsumi, T. Improvement of rutin content in buckwheat flour. In *Current Advances in Buckwheat Research, Vol. I*; Matano, T., Ujihara, A., Eds.; Shinshu University Press: Matsumoto, Japan, 1995; pp 365–372.
- Oomah, B. D.; Campbell, C. G.; Mazza, G. Effects of cultivar and environment on phenolic acids in buckwheat. *Euphytica*, in press.
- Patil, B. S.; Pike, L. M.; Hamilton, B. K. Changes in quercetin concentration in onion (*Allium cepa* L.) owing to location, growth stage and soil type. *New Phytol.* **1995**, *130*, 340–355.
- Pisha, E.; Pezzuto, J. M. Fruits and vegetables containing compounds that demonstrate pharmacological activity in humans. In *Economic and Medical Plant Research, Vol. 6*; Wagner, H., Hikino, H., Farnsworth, N. R., Eds.; Academic Press: London, UK, 1994; pp 189–233.
- Pratt, D. E. Natural antioxidants from plant material. In *Phenolic Compounds in Food and their Effect on Health II: Antioxidants and Cancer Prevention*; Huang, M-T., Ho, C-T., Lee, C. Y., Eds.; American Chemical Society: Washington, DC, 1992; pp 54–71.
- SAS Institute, Inc. SAS/STAT User's Guide, Version 6, 4th ed.; SAS Institute: Cary, NC, 1990.
- Terada, H.; Miyabe, M. Determination of rutin and quercetin in processed foods by fast semi-micro high performance liquid chromatography. *J. Food Hyg. Soc. Jpn.* **1993**, *34*, 385–391.
- Yildizoglu-Ari, N.; Altan, V. M.; Altinkurt, O.; Ozturk, Y. Pharmacological effects of rutin. *Phytotherapy Res.* **1991**, *5*, 19–23.

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