# Isoflavone Content Among Maturity Group 0 to II Soybeans

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**ABSTRACT:** This study reports the isoflavone contents of 210 soybean cultivars grown in South Dakota and explores possible relations between isoflavone contents and agronomic characteristics. Total isoflavone contents (normalized) ranged from 1161 to 2743 µg/g. A number of agronomic characteristics were documented for each variety including maturity group, hilum color, disease resistance, seed weight, yield, maturity (in days), and plant height. Varieties in maturity group I had significantly higher total isoflavones when compared to maturity group 0. Hilum color was related to differences in genistin, daidzein, and genistein content. No differences in isoflavone content were observed based on disease resistance profiles. Genistein content was found to be negatively correlated with yield, days of maturity, and plant height. Weak but significant correlations also existed between these agronomic characteristics and other isoflavones.

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**KEY WORDS:** Agronomic characteristics, daidzein, genistein, *Glycine max* L. Merrill, glycitein, HPLC, isoflavone.

Isoflavones exert a number of biological influences. Barnes *et al.* (1) documented that rats consuming a soy-based diet developed fewer mammary tumors following administration of the carcinogens *N*-methylnitrosourea and 7,12-dimethylbenz[a]-anthracene than rats fed isonitrogenous and isocaloric diets without soy. The anticarcinogenic effect may have been due to isoflavones. Isoflavones exhibit other influences such as a reduction in risk factors associated with heart disease. Soy protein products, especially those with higher concentrations of isoflavones, were shown to be hypocholesterolemic in human clinical trials (2).

Isoflavone content varies substantially among soybeans according to variety and growth conditions (3,4). Wang and Murphy (3) reported that the effect of crop year had a greater impact on the isoflavone content than did location, possibly due to climate conditions. Tsukamoto *et al.* (4) documented significant isoflavone decreases in the seeds of soybean varieties grown at a high temperature. Other investigators have documented differences in isoflavone content relating to seed physiology. Among the three major parts of the seed, the hypocotyl was found to have the highest concentration of isoflavones, at about 10 to 20% (w/w) of total seed isoflavones (3–5). In soybean seedlings, leaf flavonoid concentrations are increased by ultraviolet (UV)-B radiation and decreased soil phosphorus (6–8). The relation between isoflavone content and agronomic properties is valuable to soybean breeders for the selection of varieties that are high in individual and total isoflavones. To date, little work has been done on the relation between isoflavone content and agronomic soybean characteristics. The objective of this study was to determine useful associations that may exist between isoflavone concentrations and agronomic characteristics of soybean varieties adapted for growth in the northern United States.

## MATERIALS AND METHODS

The soybean sample set (n = 210) was collected from 1994 Crop Performance Tests. The soybean varieties were grown at the South Dakota State University (SDSU) Agronomy Farm in Brookings, South Dakota. The sample set consisted of 41 varieties in maturity group 0, 96 varieties in maturity group I, and 73 varieties in maturity group II. Hilum color and seed mass were determined for each variety. Hilum color among varieties was yellow, green, black, or brown. Yield, days of maturity, plant height, and disease resistance to three strains of *Phytophthora* (root rot) were obtained from the 1994 Crop Performance Tests (9).

Extraction procedure. Soybeans were ground with a Retsch Ultra-Centrifugal Mill Model ZM-1 (Haan, Germany) with a 0.5-mm sieve. One gram of soy meal was added to 6 mL of 80% methanol, and the mixture was stirred for 30 min. The soy meal/methanol mixture was filtered into a 50-mL volumetric flask through Whatman #42 filter paper followed by two washes with 5 mL of 80% methanol, and 80% methanol was used to dilute the filtrate to volume. Extraction conditions (stirring time, number of washes) were optimized by using the soybean variety, Mustang M-1000. The recovery rates for genistein and daidzein were consistently greater than 90%. The coefficients of variance (CV) for intraday and interday determinations were all below 2%. Although inadequate for extraction of acetyl forms of isoflavones (Murphy, P.A., personal communication), the 80% methanol extraction procedure worked well with raw soybeans, as they contained a very low amount of these acetyl derivatives.

Determination of isoflavone content. High-performance

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liquid chromatography (HPLC) was performed with a YMC-Pack ODS-AQ 303 column (5  $\mu$ m, 25 × 4.6 mm i.d.) (Wilmington, NC). A modified gradient program (10) was used, where solvent A (0.1% glacial acetic acid in water) was decreased from 85 to 69% over 45 min, while solvent B (0.1% glacial acetic acid in acetonitrile) was increased from 15 to 31%. A flow rate of 1.7 mL/min was used. The detecting UV wavelength was 254 nm.

Genistein, daidzein, and genistin peaks on sample chromatograms (Fig. 1) were confirmed using standards (Sigma Chemical Company, St. Louis, MO). The remaining isoflavones, daidzin, glycitin, malonyl daidzin, malonyl glycitin, malonyl genistin, acetyl daidzin and acetyl genistin, were identified by liquid chromatography-mass spectroscopy (LC-MS), since standards for these seven isoflavones were not commercially available. The LC-MS procedure was performed on the Mustang M-1000. Concentrations of the isoflavones were calculated from standard curves and expressed as micrograms per gram of dry weight. Equations for calculating genistein, daidzein, and genistin content were developed using known concentrations of the standards and their respective peak areas. Concentrations of malonyl genistin, acetyl genistin, and malonyl glycitin were calculated based on the standard curve of genistin with the adjustment of molecular weight ratio. Malonyl daidzin, acetyl daidzin, and daidzin concentrations were calculated from a standard curve with daidzein. Although there were no analytical standards for these isoflavones, the UV absorption per mole of isoflavones was identical (11). All isoflavone concentrations were reported as normalized and on a dry mass basis. Moisture content of the soybean varieties was determined following AOAC Method 925.10 (12), except 1 g of soy meal was used rather than 2 g.

Statistical analysis. Statistical analyses were performed using the SAS software package (13). Differences in individual and total isoflavone content among soybean varieties were determined using PROC ANOVA, with the least significant difference test. PROC GLM was used to examine differences in individual and total isoflavone content among maturity



**FIG. 1.** A typical high performance liquid chromatogram of isoflavones in soybeans. Peaks are identified as follows: 1 = daidzin, 2 = glycitin, 3 = genistin, 4 = malonyl daidzin, 5 = malonyl glycitin, 6 = malonyl genistin, 7 = daidzein, 8 = acetyl genistin, 9 = genistein.

groups, hilum color groups, and disease resistance designations. PROC CORR tested potential relations of individual and total isoflavone contents with seed weight, days of maturity, yield, and plant height. Pearson's r was reported from PROC CORR as an indicator of the strength and the direction of these relationships. Relations between these variables and individual and total isoflavone content were considered significant at P < 0.05.

## **RESULTS AND DISCUSSION**

HPLC analysis detected 10 isoflavones including: daidzin, glycitin, genistin, malonyl daidzin, malonyl glycitin, malonyl genistin, daidzein, acetyl genistin, acetyl daidzin, and genistein. Glycitein and acetyl glycitin were not quantified due to low concentrations in these soybean samples.

The isoflavone content of individual soybean varieties is listed in Table 1. The values for total genistein, total daidzein, and total glycitein represent the summation of all of the free and conjugated forms. Differences in isoflavone content were observed between soybean varieties. The five soybean varieties that yielded the highest total isoflavone content were Asgrow A-1395 (2743 µg/g), Prairie Brand PB-137 (2539 µg/g), Stine 2500 (2528 µg/g), Prairie Brand PB-193 (2526 µg/g), and Golden Harvest H-1196 (2520 µg/g). The overall range in total isoflavone content was 1116 to 2743 µg/g. Similar observations were made among other soybean varieties (3–5,14).

Significant differences in individual and total isoflavone content were observed among maturity groups. Maturity group I had significantly higher total isoflavones when compared to maturity group 0 (Table 2). However, the differences between groups I and II and groups II and 0 were not significant. Genistein content in group II varieties was significantly lower than in groups 0 and I. However, daidzein content in group I varieties was higher than in the other two groups. Genistin content was highest in maturity group 0. Maturity group 0 yielded significantly lower malonyl daidzin content than maturity groups I and II. Malonyl genistin content was significantly higher in maturity group I than in maturity group 0. Daidzin, glycitin, malonyl glycitin, and acetyl genistin content did not differ significantly among the three maturity groups.

Maturity group was not considered as a variable in previous studies. It is evident, from the results presented in this study, that the isoflavone composition of samples varies among maturity groups. The underlying mechanism for these observations has not been determined.

Isoflavone content also varied according to hilum color (Table 3). Genistin content was significantly higher in varieties with a green hilum when compared to varieties having black and brown hilums. Soybean varieties with black hilums contained lower levels of daidzein than varieties with yellow hilums. Genistein content was lowest in varieties having black hilums. Total isoflavone content did not differ significantly among hilum color groups. Although differences in

TABLE 1	
Isoflavone Contents (normalized) of Soybean Varieties Grown in Brookings, South Dakota	

		Total	Total <sup>a</sup>	Total <sup>a</sup>	Total <sup>a</sup>			Total	Total <sup>a</sup>	Total <sup>a</sup>	Total <sup>a</sup>
Variety	Maturity	isoflavones (ua/a)	genistein (%)	daidzein (%)	glycitein (%)	Variety	Maturity	isoflavones (ua/a)	genistein (%)	daidzein (%)	glycitein (%)
Mustang M-1000	0	2162.8	60	34	6	Latham 170		2384.0	56	39	7
Mustang M-1050	0	2202.5	61	34	7	Ehrich D-1398	Ì	1983.7	51	42	9
Mustang M-1040	0	1932.2	62	31	8	Ehrich D-1933	I	1927.4	56	36	10
Mustang M-0770	0	1527.3	58 58	34	10	Dairyland DSR-173		2038.9	58 57	35	10
Mustang E-0050 Mustang E-0990	0	2221.2	59	36	7	Top TF1406	I	2515.5	55	40	5
Sexauer SX0832	0	1912.9	61	33	8	Top TF1334	I	1857.7	59	34	8
Sexauer 0494X	0	1689.7	55	35	11	Kaltenberg KB171	I	2367.4	58	37	7
Sexauer SX0332 Pioneer 9071	0	1961.3	57 60	37 34	9	Kallenberg KB151 Kaltenberg KB154	I	2407.7	57	37 40	7
Pioneer 9092	Ő	2520.3	58	37	6	Kaltenberg KB162	i	2004.0	58	36	7
Dekalb CX0996	0	2021.3	61	33	8	Kaltenberg KB174	I.	2332.3	56	39	6
Kruger K0999	0	2470.8 2100 5	60 55	34	10	Ciba 3103 Ciba 2144		2364.6	57	38	6
Kruger K0909A	0	2147.6	64	30	10	Pavco 9319	I	2318.4	57	38	7
Kruger K0909	0	2287.2	61	33	8	Payco 9419	I	2339.3	58	37	6
Arrowhead 8450	0	2052.8	61	33	8	Legend LS1994	I	1973.9	57	38	6
Dvna-Gro 3033	0	1815.3	60 59	33	9 11	Terra TS194F	I	2297.8 2345.1	54 56	4 I 39	6
Ziller BT1330	0	2000.7	60	33	7	Terra TS195E	İ	1501.2	52	37	12
Ziller S74	0	1671.4	55	37	10	Golden					
McCall-00-CK	0	2212.6	62	33	7	Harvest H-1196	I	2522.0	56	34	7
Dassel	0	2010.5	59 61	30	8 9	Harvest H-1140	I.	2103 7	55	30	7
Dawson-0-CK	Ő	1952.0	60	34	8	Prairie Brand PB-193	i	2528.3	57	38	6
Evans	0	1575.5	63	30	9	Prairie Brand PB-137	I.	2541.5	57	38	7
Glenwood	0	1465.4	61	32	9	ICI D162	I	2283.3	59	35	8
Lambert	0	1363.4	62	30	o 9	ICI D138	I	1502.5	57	30 41	10
Ozzie	0	1497.0	57	36	9	Profiseed PS1504	Ì	2197.6	54	40	6
Simpson	0	1794.4	60	34	8	Profiseed PSX217B	I	2139.4	53	41	7
Parker-I-CK	0	1/49.4	62 57	30	9 10	Profiseed PS2305		21/1.2	55 55	39	/
SL92-1764M	0	1689.6	59	35	8	Great Lakes GL1927	I	1648.1	56	37	9
SL92-1233M	0	2119.4	58	33	8	Dyna-Gro 3038	I	1819.8	55	38	8
SL82-1272M	0	2062.7	56	38	9	Renze R1994	I.	1751.3	56	37	8
SL92-1323IVI SL92-1357M	0	1/51.1	59 62	34 32	8 8	Ziller BT1422	I	2170.8	59 59	30 35	6 8
SL92-1401M	Õ	1996.2	56	33	10	Ziller BT2772	i	2370.1	59	37	5
SL92-1461M	0	1733.3	59	34	11	Premier P-2012	I.	2065.7	62	33	7
ND88597	0	1736.4	57	36	8	Premier P-1970	I	2402.4	61	32	7
Asgrow A1900	1	2745.7	60	34	7	Dawson-0-CK	I	1929.4	60	40 34	9
Mustang M-1140	I	2465.0	56	40	6	Alpha	I	1898.4	59	32	10
Mustang M-1170		1595.2	56	38	9	Bell-SCN-CK	I.	1463.5	56	36	12
Mustang M-1122		2423.2 2264.6	56 58	38 37	6 7	BSR 101	I	1976.8	57 58	37 37	8
Northrup King S19-90		2210.6	58	37	7	Hardin	İ	1778.6	60	33	9
Northrup King S12-22	I	2040.9	59	35	7	Kasota	I	1439.3	61	32	10
Northrup King S16-60		2037.4	52	42	7	Kato	I	1946.4	60	34	7
Sexauer SX1232	1	2404.9 1947 5	53 58	42	9	Leslie Parker-I-CK	I	1970.1 1949-2	58 60	35 31	8 10
Pioneer 9111	i	2167.9	57	39	6	Sibley	i	1965.9	57	38	9
Pioneer 9162	I	1708.2	59	32	9	Sturdy-II-CK	I.	2209.3	55	39	7
Pioneer 91/1 Pioneer 91/1		1936.9	58 57	33	11	SL92-1194M SL92-1170M		2047.4	57	37	/
Dekalb CX117		2091.1	60	30	9	SL92-1201M	I I	1982.8	58	35	8
Dekalb CX121	Ì	2412.3	58	37	8	SL92-1207M	Ì	1496.9	57	35	10
Agripro AP1347	1	2133.0	53	42	7	SL92-1328M	I	1521.6	60	33	9
Agripro AP1880 Sands SO1113	1	1697.0	57	35	10	SL92-1362M SL92-1412M		1951.3	56 55	38	8
Desoy 1777	i	1997.8	60	33	8	SL92-2844M	i	1668.4	59	35	9
Desoy 1707	I	2448.1	54	38	9	M97642	I	1810.6	58	37	8
Desoy D-1819		2319.6	53	42	9	Filler Parker		1876.3	60	31	9
Desoy D-1909 Desoy D-1999	1	2123.3	55	42 39	7	Asgrow A2242 Asgrow A2012	11	2322.1	59	32 36	9
Kruger K1444	i	1990.9	51	44	8	Mustang M-1200	II II	2311.7	53	39	8
Kruger K1717+	1	2105.4	55	40	7	Mustang M-1222	11	1810.3	57	34	12
Kruger K1818+		2029.2	63	32	7	Mustang E-2215		2217.7	56	36	10
Kruger K1313+		2223.5	57	38	7	Pioneer 9241	11	2082.9	54	41	6
Kruger K1716	I	1982.9	56	36	9	Pioneer 9252	II	1680.4	54	40	8
Arrowhead 8600	ļ	2113.2	57	37	9	Pioneer 9204	11	1511.3	55	35	11
Arrownead 8495		2024.9	55 58	39	8 7	Dekalb CX232		2033.9	65 55	30	8
	I	2003.7	00	30	/	Denain CA220	11	1070.0	00	57	7
(continued)											

#### TABLE 1 (continued)

		Total	Total <sup>a</sup>	Total <sup>a</sup>	Total <sup>a</sup>			Total	Total <sup>a</sup>	Total <sup>a</sup>	Total <sup>a</sup>
		isoflavones	genistein	daidzein	glycitein			isoflavones	genistein	daidzein	glycitein
Variety	Maturity	(µg/g)	(%)	(%)	(%)	Variety	Maturity	(µg/g)	(%)	(%)	(%)
Sands SQI214		2149.8	53	38	10	Prairie Brand 227EXF	>	1924.2	49	44	10
Sands SQI230	11	2476.8	63	33	7	ICI D213	П	2501.7	63	33	6
Hy Vigor 2050	11	2117.9	53	39	8	Profiseed PS2555	П	1821.4	63	32	6
Desoy 2333	11	1493.1	54	39	12	Profiseed PS2224	11	2275.6	52	41	7
Desoy D2020+	11	2185.8	52	39	9	Profiseed PS2134	11	2224.3	55	39	8
Desoy D2162+	11	2150.0	53	40	11	Star EXP9321B	11	1797.6	55	36	10
Desoy D2121	11	2121.3	52	40	10	Star EXP9321	11	1964.5	55	38	9
Desoy D2303+	11	1886.9	57	37	10	Great Lakes GL2237	11	1802.5	54	40	8
Desoy D2303	11	2033.4	51	43	8	Great Lakes GL2405	П	1944.3	56	38	8
Kruger K2162	11	1759.4	56	35	12	Renze R2302	П	2161.6	53	39	9
Kruger K2525	11	1980.6	64	31	9	Renze R2395	11	1703.7	55	38	10
Kruger K2021	11	2312.5	54	39	7	Premier P-2122E	П	2301.9	64	31	5
Kruger K2020	11	2050.8	52	43	8	Parker-I-CK 1032	11	1633.3	60	31	9
Kruger K2222	11	2147.1	64	31	6	Century 84	11	2139.8	56	38	8
Kruger K2323+	11	2067.9	55	39	7	Conrad 1031	11	1622.7	57	36	9
Enrich E-298	11	2122.1	53	39	9	Conrad BC 1027	11	1491.9	55	38	9
Dairyland DSR217	11	2261.2	53	39	10	Corsoy 79	11	1598.0	56	37	8
Dairyland DSR222	11	2457.8	55	39	8	Holt 1040	11	2204.5	61	32	8
Stine 2500	11	2530.2	58	38	6	Kenwood 1048	11	1755.0	55	37	10
Sansgaard S-201	11	2125.9	56	38	7	Kenwood BC	11	1917.1	56	38	8
Ciba 3202	11	1637.7	57	37	9	Marcus 1052	П	2022.0	57	33	10
Payco 9023	11	2166.8	53	39	8	Newton 1006	11	1985.9	65	29	10
Payco 9225	11	1863.1	64	31	9	Sturdy-II-CK 1007	П	2130.0	56	39	7
Payco 9421	11	2093.6	56	37	7	Resnik-III-CK 1002	П	1572.3	54	35	11
Terra TS253	11	1965.0	63	32	7	SL92-1153M	11	1959.3	58	35	9
Terra TS294E	11	1842.7	52	42	7	SL92-1174M	П	1792.2	56	38	8
Terra TS225E	11	1946.2	65	30	7	SL92-1205M	11	1935.4	56	38	7
Golden						SL92-1225M	11	1620.4	54	40	8
Harvest H-1263	11	1918.1	65	30	6	SL92-1227M	11	1630.8	57	37	8
Golden						SL91-1252N	П	1821.3	55	38	8
Harvest H-1228	11	1161.7	54	38	10	SL91-1657N	11	2016.5	56	37	8
Prairie Brand PB-214	11	2311.6	53	40	7	IA2008	11	1605.9	56	35	10

<sup>a</sup>The values for total genistein, total daidzein, and total glycitein represent the summation of all free and conjugated forms.

content were observed for individual isoflavones on the basis of hilum color, no discernible trend was found that could provide a suitable explanation for these findings.

Past research has demonstrated that isoflavones can function as fungicides (15). To test this premise, the relation between disease resistance to three strains of *Phytophthora* (root rot) and isoflavone content was investigated for the soybean sample set. No differences in isoflavone content were detected for susceptible, unknown, and resistant varieties.

A weak, but significant, negative correlation was observed between seed mass and the malonyl glycitin content of the samples (r = -0.18). Correlations between seed weight and the other isoflavones were insignificant. This indicated that seed weight was a poor indicator for isoflavone content of the sample.

Yield (bushels/acre) for each variety was obtained from the 1994 Crop Performance Tests. Weak, but significant, correlations were observed between yield and daidzin (r = 0.19), glycitin (r = 0.20), malonyl daidzin (r = 0.36), malonyl glycitin (r = 0.21), malonyl genistin (r = 0.16), daidzein (r = -0.20), acetyl daidzin (r = 0.21), and total isoflavone content (r = 0.20). A stronger correlation was observed between yield and genistein (r = -0.44). This indicated that high-yield varieties tended to have lower genistein concentration. Yield was not found to be correlated significantly with genistin and acetyl genistin.

The number of days required for seed maturity was recorded for each variety in the soybean sample set as part of the 1994 Crop Performance Tests. This differs from maturity groups, which are categorical designations for the number of days required for seed maturity. Days of maturity yielded insignificant correlations with daidzin, glycitin, malonyl glycitin, malonyl genistin, acetyl genistin, and total isoflavone content. Weak, but significant, linear correlations were ob-

TABLE 2	
Comparison of Isoflavone Composition Among Maturity C	Groups <sup>a,b</sup>

	No. of				Malonyl	Malonyl	Malonyl		Acetyl	Acetyl		
Maturity	observations	Daidzin	Glycitin	Genistin	daidzin	glycitin	genistin	Daidzein	genistin	daidzin	Genistein	Total
0	41	140.6 <sup>a</sup>	47.5 <sup>a</sup>	232.0 <sup>a</sup>	397.9 <sup>b</sup>	78.2 <sup>a</sup>	829.1 <sup>b</sup>	17.8 <sup>b</sup>	28.0 <sup>a</sup>	85.8 <sup>b</sup>	38.8 <sup>a</sup>	1891.1 <sup>b</sup>
I	96	145.3 <sup>a</sup>	47.6 <sup>a</sup>	211.0 <sup>b</sup>	397.5 <sup>a</sup>	79.1 <sup>a</sup>	897.9 <sup>a</sup>	31.9 <sup>a</sup>	28.1 <sup>a</sup>	92.3 <sup>a</sup>	37.5 <sup>a</sup>	2068.4 <sup>a</sup>
11	73	141.1 <sup>a</sup>	49.1 <sup>a</sup>	210.4 <sup>b</sup>	477.8 <sup>a</sup>	82.8 <sup>a</sup>	864.4 <sup>a,b</sup>	13.9 <sup>b</sup>	28.3 <sup>a</sup>	91.2 <sup>a</sup>	16.7 <sup>b</sup>	1976.1 <sup>a,b</sup>
LSD <sup>a</sup>	_	9.1	4.3	14.9	31.3	5.7	45.6	5.4	0.9	3.3	5.2	100.5

<sup>a</sup>Values with the same letter, in the same column, are not significantly different (P < 0.05). LSD, least significant difference (P < 0.05). <sup>b</sup>Isoflavone contents are expressed in µg/g.

TABLE 3			
Relation of	Isoflavone	Content to Hilum Color of Soybean Varieties	a,b
L P.L	NI 6	Malaws d	N A = L = ve v d

Hilum	No. of				Malonyl	Malonyl	Malonyl		Acetyl	Acetyl		
color	observations	Daidzin	Glycitin	Genistin	daidzin	glycitin	genistin	Daidzein	genistin	daidzin	Genistein	Total
Green	10	150.1 <sup>a</sup>	47.9 <sup>a</sup>	239.3 <sup>a</sup>	478.8 <sup>a</sup>	83.8 <sup>a</sup>	908.8 <sup>a</sup>	24.9 <sup>a,b</sup>	28.0 <sup>a</sup>	91.5 <sup>a</sup>	36.2 <sup>a</sup>	2089.2 <sup>a</sup>
Yellow	57	142.7 <sup>a</sup>	47.7 <sup>a</sup>	220.6 <sup>a,b</sup>	452.5 <sup>a</sup>	77.2 <sup>a</sup>	870.7 <sup>a</sup>	28.4 <sup>a</sup>	27.9 <sup>a</sup>	90.1 <sup>a</sup>	39.5 <sup>a</sup>	1997.5 <sup>a</sup>
Black	80	144.0 <sup>a</sup>	47.1 <sup>a</sup>	209.8 <sup>b</sup>	488.2 <sup>a</sup>	78.4 <sup>a</sup>	872.9 <sup>a</sup>	17.3 <sup>b</sup>	28.4 <sup>a</sup>	91.2 <sup>a</sup>	22.2 <sup>b</sup>	1999.8 <sup>a</sup>
Brown	63	140.5 <sup>a</sup>	49.9 <sup>a</sup>	212.3 <sup>b</sup>	465.3 <sup>a</sup>	84.6 <sup>a</sup>	868.7 <sup>a</sup>	24.8 <sup>a,b</sup>	28.2 <sup>a</sup>	90.3 <sup>a</sup>	32.1 <sup>a</sup>	1997.0 <sup>a</sup>
LSD/0.05	_	13.8	6.4	22.7	50.6	8.5	69.9	9.0	1.4	5.2	8.7	155.2

<sup>*a,b*</sup>For footnotes see Table 2.

served for genistin (r = -0.27), malonyl daidzin (r = 0.17), and daidzein (r = -0.33). For genistein, days of maturity yielded the strongest linear correlation (r = -0.59), indicating that maturity of seeds has a negative relationship with genistein content in soybean seeds. This follows the results from maturity group comparisons in that longer maturity resulted in a reduction of genistein content.

Plant height was measured from the soil surface to the top node of the main stem for the 1994 Crop Performance Tests. Weak, but significant, correlations were observed between plant height and genistin (r = -0.29), malonyl daidzin (r =0.15), or daidzein (r = -0.23). Insignificant linear correlations were found between plant height and daidzin, glycitin, malonyl glycitin, malonyl genistin, acetyl genistin, and total isoflavones. The strongest linear correlation was observed for genistein (r = -0.46). This indicated that taller plants tend to have lower genistein content. Similar to yield, plant height was found to be a poor indicator of total isoflavone content. This has also been confirmed by Tsukamoto et al. (4). However, compared to other isoflavones, plant height, yield, and maturity were better indicators of genistein content. Environmental factors such as crop year, location, rainfall, and temperature apparently influence the relations observed between agronomic seed characteristics and isoflavone content.

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## REFERENCES

- Barnes, S., C. Grubbs, K.D. Setchell, and J. Carlson, Soybeans Inhibit Mammary Tumors in Models of Breast Cancer, *Prog. Clin. Biol. Res.* 347:239–253 (1990).
- 2. Potter, S. M., J. Baum, P. Surya, and J.W. Erdman, Effects of Soy Protein and Isoflavones on Plasma Lipid Profiles in Post-

menopausal Women, Second International Symposium on the Role of Soy in Preventing and Treating Chronic Disease, edited by M. Messina, Brussels, Belgium, 1996, pp. 22.

- Wang, H.J., and P.A. Murphy, Isoflavone Composition of American and Japanese Soybeans in Iowa: Effects of Variety, Crop Year, and Location, J. Agric. Food. Chem. 42:1674–1677 (1994).
- Tsukamoto, C., S. Shimadu, K. Igita, S. Kudou, M. Kokuun, K. Okuibo, and K. Kitamura, Factors Affecting Isoflavone Content in Soybean Seeds: Changes in Isoflavones, Saponins, and Composition of Fatty Acids at Different Temperatures During Seed Development. *Ibid.* 43:1184–1192 (1995).
- 5. Eldridge, A.C., and W.F. Kwolek, Soybean Isoflavones: Effect of Environment and Variety on Composition, *Ibid.* 31:394–396 (1983).
- Graham, T.L., Flavonoid and Isoflavonoid Distribution in Developing Soybean Seedling Tissues and in Seed and Root Exudates, *Plant Physiol.* 95:594–603 (1991).
- 7. Buttery, B.R., and R.I. Buzzell, Varietal Differences in Leaf Flavonoids of Soybeans, *Crop. Sci.* 13:103–106 (1973).
- Murali, N.S., and A.H. Teramura, Effects of Ultraviolet-B Irradiance on Soybean. VI. Influence of Phosphorus Nutrition on Growth and Flavonoid Content, *Physiol. Plant.* 63:413–416 (1985).
- Hall, R.G., and P.D. Evenson, Soybeans: 1995 Variety Recommendations (1994 Crop Performance Test Results), SDSU EC 775, South Dakota State University, Brookings, South Dakota, 1995.
- Wang, H.J., and P.A. Murphy, Isoflavone Content in Commercial Soybean Foods, J. Agric. Food Chem. 42:1666–1673 (1994).
- Kuduo, S., Y. Fleury, D. Welti, D. Magnolato, T. Uchida, K. Kitamura, and K. Okubo, Manonyl Isoflavone Glycosides in Soybean Seeds (*Glycine max* L. Merrill), *Agric. Biol. Chem.* 55: 2227–2233 (1991).
- 12. Association of Official Analytical Chemists, *Official Methods* of Analysis of the Association of Official Analytical Chemists, 15th edn., Arlington, 1990.
- 13. SAS Institute, Inc., SAS/STAT Guide for Personal Computers, 6th edn., Cary, North Carolina, 1987.
- Fleury, Y., D.H. Welti, G. Philippossian, and D. Magnolato, Soybean (malonyl) Isoflavones: Characterization and Antioxidant Properties, ACS Symposium Series 507:98–113 (1992).
- Naim, M., B. Gestetner, S. Zilkah, Y. Birk, and A. Bondi, Soybean Isoflavones, Characterization, Determination, and Antifungal Activity, J. Agric. Food. Chem. 22:806–810 (1974).

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