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Comparison of Compositions of Imported Genetically Modified and Organic Soybeans Purchased from Taiwan Market

Huan-Yu Lin[1], Bo-Chou Chen[1], Mei-Li Chao[1], Hui-Wen Chang[1], Hsin-Tang Lin[2], Wen-Shen Chu[1,*]

[1]Bioresource Collection and Research Center, Food Industry Research and Development Institute, Hsinchu 300, Taiwan
[2]Food and Drug Administration, Ministry of Health and Welfare, Taipei 115, Taiwan
(Present address: Graduate Institute of Food Safety, National Chung Hsing University, Taichung 402, Taiwan)
*Corresponding author: cws@firdi.org.tw

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Abstract Soybean is an excellent source of plant-derived protein, and soy products are one of the important protein sources for vegetarians in Taiwan. Soybean production in Taiwan is not self-sufficient. Taiwan imports 2.5 million tons of soybeans annually. More than 90% of the imported soybeans are genetically modified. To provide an objective assessment on safety of genetically modified soybean and for post-market monitoring, we conducted a comparative assessment on key component compositions between imported genetically modified and organic soybeans. All the soybean samples were purchased from the local market to simulate the status of Taiwanese consumers purchasing soybeans. The genetically modified soybean samples were herbicide-tolerant soybeans. The contents of the proximate, the amino acid composition, the fatty acid composition, vitamins, minerals, isoflavones, and anti-nutritional factors of soybean samples were analyzed. Most contents of the key components of the genetically modified soybean had no significant difference with those of the organic soybean in this study. However, the contents of cysteine and α-linoleic acid were significantly lower in the genetically modified soybean samples, and the contents of crude fat, palmitic acid, stearic acid, oleic acid, linoleic acid, vitamin K1, and calcium were significantly higher in the genetically modified soybean samples. But they were all within the range of reference values. A total of 314 pesticide residues in each of the samples were analyzed. Only glyphosate residue was detected in the genetically modified soybean samples, but it is well below the threshold prescribed by the government. In summary, the imported genetically modified and organic soybeans purchased from Taiwan market were shown to be substantially equivalent in composition.

Keywords: *genetically modified soybean, organic soybean, composition, substantially equivalent*

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1. Introduction

As the world population continues to increase, it will be accompanied by an increased demand for food. The commercialization of biotech crops, also known as genetically modified crops, increases productivity significantly and reduces production costs substantially. In 2018, genetically modified crops were grown on an area of 191.7 million hectares. Genetically modified soybeans are the most planted genetically modified crops, with 95.9 million hectares grown worldwide. Genetically modified soybeans are planted in 10 countries, and two of the major countries are the United States (34.08 million hectares) and Brazil (34.86 million hectares). In the United States, the adoption rate of genetically modified soybean is 93.3% [1].

Soybean is an important protein source, especially for vegetarians. Processed soybean products are daily consumed foods in Taiwan, such as soymilk, tofu, dried tofu and bean cream skim. Soybean consumption in Taiwan was 56.91 g/person/day [2], almost 57 fold higher than that in western countries [3]. Soybean production in Taiwan is not self-sufficient. Most soybean grains are imported from abroad. Over the past decade, more than two million metric tons of soybeans are imported into Taiwan each year. In 2018, soybean imports in Taiwan were 2.63 million metric tons, of which 2.55 million metric tons were genetically modified soybeans, accounting for 96.94% [4].

According to the World Health Organization (WHO), the Food and Agriculture Organization (FAO) and the Organization for Economic Co-operation and Development (OECD) on the safety assessment of genetically modified foods, the policies should be constructed on molecular, biological and chemical properties of food. Safety assessment of genetically modified food crops is based on the concept of substantial equivalence, comparing genetically modified food with a similar traditional food that has been proven to be safe during normal use [5].

Genetically modified foods have undergone various safety assessments before sell. Up to the end of Sep. 2019, a total of 149 genetically modified events have been approved for food use by the Taiwan Food and Drug Administration (TFDA), including 28 soybean events, 81 maize events, 28 cotton events, 11 canola events and 1 sugar beet event [6]. However, some consumers still have doubt about the objectivity of the safety assessment information provided by the industries. To eliminate public safety concerns, we conducted an objective assessment on the safety of soybeans placed on the market. The contents of proximate, amino acids, fatty acids, vitamins, minerals, isoflavones, and anti-nutritional factors of genetically modified and organic soybean samples purchased from Taiwan local markets in 2016 were analyzed. Although some significant differences in composition were found, all the contents were within the reference range. The glyphosate residue was only detected in genetically modified soybean samples. But the residue level was well below the threshold prescribed by the government.

2. Materials and Methods

2.1. Soybean Samples

A total of 20 soybeans samples, including 10 of each genetically modified and organic soybean samples, were purchased from local markets in Taiwan. The genetically modified soybeans were imported from the United States in 2015 (2016GM), and the organic soybeans were grown in the United States, Canada and Brazil in 2015 (2016OS). The soybean samples in each category were divided into three parts for compositional analysis, including proximate, amino acids, fatty acids, vitamins, minerals, isoflavones, and anti-nutritional factors.

2.2. DNA Isolation and PCR Assay

The soybeans were homogenized directly in a blender. 100 mg of soybean powders were collected for DNA isolation by using the DNeasy® plant mini kit (Qiagen, Valencia, CA), following the manufacturer's instructions. The PCR primers were designed for the CaMV35S promoter (P35S), the CaMV35S terminator (T35S), the *nos*-terminator of *Agrobacterium tumefaciens* strain CP4 (T*nos*), the gene encoding 5-enolpyruvulshikimate-3-phosphate synthase (EPSPS) of *A. tumefaciens* stain CP4 (*cp4 epsps*), the pat gene derived from *Streptomyces viridochromogenes*, the *cry1Ab/cry1Ac* gene derived from *Bacillus thuringiensis* subsp. Kurstaki strain HD73 (*cry1Ab/Ac*), the *dmo* gene derived from *Stenotrophomonas maltophilia* strain DI-6 (*dmo*), the *gm-hra* gene derived from *Glycine max*, the *crs*1-2 derived from *Arabidopsis thaliana* (*gm-hra*), the *Pj.D6D* gene derived from *Primula juliae* (*Pj.D6D*), the *epsps* gene with double mutates from *Zea mays* (*2mepsps*), and lectin gene from *G. max*. Controls used in this study were (i) non-transgenic soybean (Ecocert Canada, 10145-14-01); and (ii) certified reference materials: A2704-12 soybean (AOCS 0707-B6), CV127 soybean (AOCS 0911-C), FG72 soybean (AOCS 0610-A2), GTS 40-3-2 soybean (ERM-BF410gk), MON87701 soybean (AOCS 0809-A), MON87708 soybean (AOCS 0311-A), MON87769 (AOCS 089-B), and 305423 soybean (ERM-BF426d). The detection limit for the tested genes was 0.1%. The PCR conditions were as previously described [7].

2.3. Proximate Analysis

Proximate analysis of the soybean samples was conducted using the standard Association of Official Analytical Chemists (AOAC) procedures [8]. Moisture content was determined by loss of weight upon drying in an oven at 135°C for 2 hr (Method No. 930.15) [8]. Ash content was determined by incineration (550°C) of known weights of the samples in a muffle furnace (Method No. 930.05) [8]. Crude fat content was determined by acid hydrolysis of known weights of the samples (Method No. 922.06) [8]. Crude protein content was determined by digestion of the samples in a block digester and determination of ammonia (Method No. 976.06) [8]. Dietary fiber analysis was carried out by enzymatic gravimetry (Method No. 985.29) [8]. Carbohydrate content was calculated as (100 [g/100 g] – moisture [g/100 g] - ash [g/100 g] - crude fat [g/100 g] - crude protein [g/100 g] – fiber [g/100 g]).

2.4. Fatty Acid and Amino Acid Analysis

The total, saturated, and unsaturated fatty acids were analyzed by hydrolytic extraction using gas chromatography (Method No. 996.06) [8]. For amino acid analyses, samples were hydrolyzed according to the methane disulfonic acid method and determined using high-performance liquid chromatography [9].

2.5. Vitamin Analysis

Vitamin B1 was analyzed by oxidizing thiamine to thiochrome and measuring fluorescence (Method No. 986.27) [8]. Vitamin B2 was analyzed using fluorescence detection (Method No. 981.15) [8]. Folic acid content was determined by fluorometry (Method No. 944.12) [8]. Vitamin E was analyzed using liquid chromatography (Method No. 2012.09) [8]. Vitamin K1 was analyzed by high-performance liquid chromatography (Method No. 2002.05) [8].

2.6. Mineral Analysis

Calcium, magnesium, potassium, phosphorus and iron were analyzed by inductively coupled plasma–optical emission spectrometry (Method No. 2011.14) [8].

2.7. Oligosaccharide, Anti-nutritional Factor and Isoflavones Analysis

Extraction of stachyose and raffinose from soybean samples was performed and determined using high-performance liquid chromatography [10]. The content of lectin was determined by enzyme-linked immunosorbent assay (ELISA). Phytic acid was analyzed by colorimetry [11]. The content of trypsin inhibitor was determined by colorimetry, according to the American Oil Chemists' Society (AOCS) official method Ba12-75 [12]. The content of isoflavones was determined by high-performance liquid chromatography. The calibration curve was constructed with external standards of daidzin, daidzein, genistin, genistein, glycitin, glycitein, malonyl daidzin, malonyl genistin, malonyl glicitin, acetyl daidzin, acetyl genistin, and acetyl glycitin [13].

2.8. Statistical Analysis

Experimental values were mean ± standard deviation (SD) from three separate experiments. Significance was assessed using student *t*-tests. The statistical level of significance was present at 0.05.

3. Results

3.1. Identity of Soybean Samples

Genetically modified or organic soybean samples were collected from Taiwan local markets according to their labeling, and evaluated to simulate Taiwan consumers' diet on soybeans. To confirm the identity of the samples, PCR assays were conducted to examine the transgenes. No transgenic genes were detected in the 2016OS sample (Table 1 and Figure 1). P35S, T35S, T*nos*, *cp4 epsps* and *pat* were detected in the 2016GM samples while *cry1Ab/Ac*, *dmo*, *gm-hra*, *csr1-2*, *PjD6D*, and *2mepsps* were not detected (data not shown). According to the approval list of the genetically modified soybean events, the 2016GM soybean samples contained the glyphosate herbicide-tolerant events [GTS 40-3-2 (Roundup Ready™ soybean, MON-Ø4Ø32-6), MON89788 (Genuity® Roundup Ready 2 Yield™, MON-89788-1), MON87705 (Vistive Gold™, MON-877Ø5-6)] and glufosinate herbicide-tolerant events [A2704-12 (Liberty Link™ soybean, ACS-GMØØ5-3), A5547-127 (Liberty Link™ soybean, ACS-GMØØ6-4)].

3.2. Proximate Composition

The contents of the proximate, the amino acid composition, the fatty acid composition, vitamins, minerals, isoflavones, and anti-nutritional factors of soybean samples were analyzed followed OECD guideline [14]. The contents of moisture, ash, crude fat, crude protein, crude fiber, and carbohydrate of the 2016GM and 2016OS soybean samples were analyzed and compared (Table 2). No significant difference was found in contents events, the 2016GM soybean sample contained the of moisture, ash, crude protein, crude fiber, and carbohydrate between the 2016OS and the 2016GM soybean samples at 5% confidence level. Significant higher content in crude fat of the 2016GM soybean sample (21.12 ± 0.25 g/100 g) was observed as compared to that of the 2016OS soybean sample (18.64 ± 0.91 g/100 g) but within the literature range of 6.97~25.0 g/100 g.

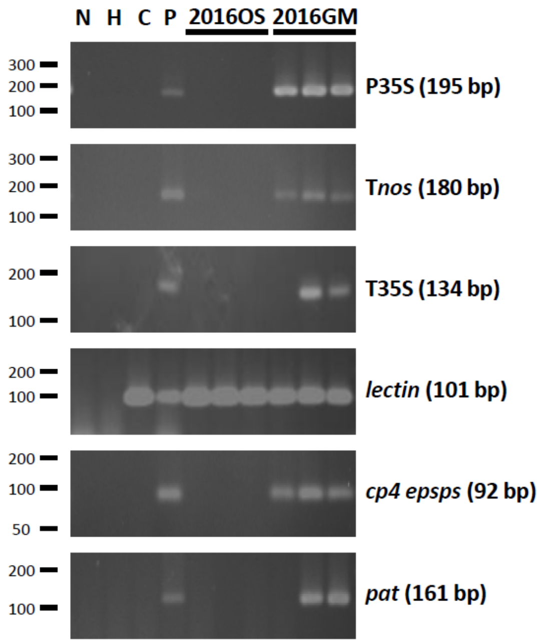

Figure 1. Screening of soybean samples for foreign transgenes. DNA was isolated from the 2016OS soybeans and 2016GM soybeans. The foreign transgenes were amplified using PCR for P35S, T*nos*, T35S, lectin gene, *cp4 epsps*, and *pat*. The detection limit is 0.1%. N, blank control; H, ddH_2O; C, non-genetically modified soybean; and P, 0.1% reference material of genetically modified soybean

Table 1. Detection of transgenes in soybean samples

Sample	P35S	T*nos*	T35S	*Cp4 epsps*	*pat*	*lectin*	Events
2016OS-1	-	-	-	-	-	+	
2016OS-2	-	-	-	-	-	+	
2016OS-3	-	-	-	-	-	+	
2016GM-1	+	+	-	+	-	+	RRS, 89788, 87705
2016GM-2	+	+	+	+	+	+	RRS, 89788, 87705, A2704-12, A5547-127
2016GM-3	+	+	+	+	+	+	RRS, 89788, 87705, A2704-12, A5547-127

3.3. Amino Acid Contents

The amino acid profile of soybean samples was investigated in this study. Seventeen amino acids of the genetically modified and the organic soybean samples were analyzed (Table 3). Most of contents for amino acids were not significantly different between the 2016OS and the 2016GM soybean samples at 5% confidence level. Significant lower content in cystine of the 2016GM soybean sample (0.31 ± 0.01 g/100 g) was observed as compared to that of the 2016OS soybean sample (0.36 ± 0.00 g/100 g). However, the contents of the 2016GM soybean samples (0.30, 0.31, 0.32 g/100g) were outside the range of 0.32~0.93 g/100 g in the ILSI Crop Composition Database [15], but within the literature range of 0.213 ~ 0.553 g/100 g [16].

3.4. Fatty Acid Contents

The fatty acid content is an indicator of chemical change in soybean oil. Forty-two fatty acids were analyzed, of which 30 were not detected. Among the fatty acid profile, those found in the highest concentrations were linoleic acid (18:2), oleic acid (18:1), palmitic acid (16:0), α-linolenic acid (18:3, n-3), and stearic acid (18:0). Comparing the 2016OS and the 2016GM soybean samples, the contents of these top five fatty acids showed significant difference, but the pattern is not synchronized. The α-linolenic acid (18:3, n-3) content was significantly lower in the 2016GM soybean sample, but the palmitic acid (16:0), the stearic acid (18:0), the oleic acid (18:1), and the linoleic acid (18:2) were significantly higher in the 2016GM soybean sample. However, these contents were all within the literature range (Table 4).

Table 2. Proximate contents of organic and genetically modified soybeans

Proximate (g/100 g)	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
Moisture	10.21 ± 0.52	9.71~10.75	10.87 ± 0.71	10.17~11.58	0.26	4.7~44.5
Ash	4.53 ± 0.02	4.30~4.67	4.49 ± 0.10	4.40~4.60	0.81	3.75~10.9
Crude fat	18.64 ± 0.91	18.03~19.69	21.12 ± 0.25	20.84~21.33	0.01*	6.97~25.0
Crude protein	36.31 ± 2.22	33.76~37.77	32.59 ± 3.48	29.48~36.35	0.19	29.51~46.6
Crude fiber	5.60 ± 0.65	4.89~6.15	5.88 ± 0.42	5.41~6.22	0.56	4.12~18.5
Carbohydrate	24.71 ± 2.96	22.96~28.12	25.05 ± 3.94	20.81~28.59	0.91	25.2~55.8

Data are mean ± SD from three separate experiments. Literature range is from ILSI Crop Composition Database v7.0, 2019. Carbohydrate content is calculated as (100 [g/100 g] – moisture [g/100 g] - ash [g/100 g] - crude fat [g/100 g] - crude protein [g/100 g] – fiber [g/100 g]). * The significant difference ($p<0.05$) is examined using *t*-test.

Table 3. Amino acid contents of organic and genetically modified soybeans

Amino Acid (g/100 g)	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
Aspartic acid	4.32 ± 0.18	4.14~4.49	4.11 ± 0.07	4.05~4.19	0.13	3.13~6.83
Threonine	1.48 ± 0.06	1.42~1.55	1.41 ± 0.03	1.38~1.45	0.19	1.07~2.177
Serine	1.86 ± 0.07	1.78~1.92	1.75 ± 0.03	1.72~1.78	0.08	0.863~2.8
Glutamic acid	6.67 ± 0.29	6.37~6.93	6.33 ± 0.12	6.23~6.46	0.13	4.35~10.9
Glycine	1.62 ± 0.06	1.57~1.68	1.56 ± 0.02	1.53~1.58	0.16	1.16~2.5467
Alanine	1.58 ± 0.06	1.52~1.64	1.51 ± 0.03	1.48~1.54	0.15	1.15~2.35
Cystine	0.36 ± 0.00	0.35~0.36	0.31 ± 0.01	0.30~0.32	0.001*	0.32~0.93
Valine	1.39 ± 0.05	1.33~1.44	1.32 ± 0.03	1.30~1.35	0.14	1.24~2.66
Methionine	0.35 ± 0.04	0.30~0.38	0.29 ± 0.03	0.27~0.33	0.14	0.285~1.15
Isoleucine	1.60 ± 0.06	1.53~1.65	1.53 ± 0.04	1.50~1.57	0.19	1.2~2.48
Leucine	2.87 ± 0.13	2.73~2.94	2.74 ± 0.06	2.68~2.79	0.17	2.04~4.13
Tyrosine	1.43 ± 0.05	1.37~1.48	1.37 ± 0.03	1.33~1.39	0.16	0.737~2.32
Phenylalanine	1.85 ± 0.09	1.75~1.91	1.76 ± 0.04	1.73~1.80	0.20	1.4~2.73
Lysine	2.42 ± 0.10	2.33~2.52	2.30 ± 0.04	2.26~2.34	0.10	1.79~3.94
Histidine	0.97 ± 0.04	0.92~1.01	0.92 ± 0.02	0.91~0.94	0.13	0.197~1.59
Arginine	2.90 ± 0.19	2.72~3.09	2.67 ± 0.04	2.64~2.72	0.11	1.73~3.926
Proline	3.00 ± 0.13	2.87~3.13	2.83 ± 0.10	2.77~2.94	0.15	1.32~2.945

Detection limit of quantification is 0.01 g/100 g. Data are mean ± SD from three separate experiments. Literature range is from ILSI Crop Composition Database v7.0, 2019. * The significant difference ($p<0.05$) is examined using *t*-test.

Table 4. Fatty acid contents of organic and genetically modified soybeans

Fatty Acid (g/100 g)	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
14:0 Myristic acid	0.01 ± 0.01	0.01~0.02	0.02 ± 0.00	0.02~0.02	0.12	0.008~0.034
16:0 Palmitic acid	2.17 ± 0.14	2.06~2.33	2.50 ± 0.04	2.46~2.53	0.02*	0.56~2.81
16:1 Palmitoleic acid	0.01 ± 0.01	0.01~0.02	0.02 ± 0.00	0.02~0.02	0.12	0.008~0.041
17:0 Margaric acid	0.02 ± 0.00	0.02~0.02	0.02 ± 0.00	0.02~0.02	-	0.010~0.031
18:0 Stearic acid	0.73 ± 0.03	0.70~0.75	0.90 ± 0.00	0.90~0.90	0.0003*	0.206~1.452
18:1 Oleic acid	3.62 ± 0.28	3.44~3.94	4.74 ± 0.06	4.68~4.80	0.002*	1.12~9.43
18:2 Linoleic acid	10.16 ± 0.51	9.83~10.75	11.15 ± 0.25	10.91~11.4	0.04*	2.51~13.23
18:3 Linolenic acid (n-3)	1.74 ± 0.04	1.72~1.79	1.57 ± 0.01	1.56~1.58	0.002*	0.32~2.081
20:0 Arachidic acid	0.06 ± 0.01	0.05~0.06	0.06 ± 0.01	0.06~0.07	0.23	0.017~0.133
20:1 Eicosenoic acid	0.02 ± 0.00	0.02~0.02	0.02 ± 0.01	0.02~0.03	0.37	0.013~0.079
22:0 Behenic acid	0.07 ± 0.01	0.07~0.08	0.08 ± 0.01	0.07~0.08	0.52	0.024~0.162
24:0 Lignoceric acid	0.02 ± 0.01	0.02~0.03	0.03 ± 0.00	0.03~0.03	0.12	0.0138~0.0699

Data converted from g/100 g to percent of total fatty acid. Total fatty acid is calculated as the sum of total saturated fatty acid, total monounsaturated fatty acid and total polysaturated fatty acids. Detection limit of quantification is 0.01 g/100 g fatty acid. Literature range is from ILSI Crop Composition Database v7.0, 2019. * The significant difference ($p<0.05$) is examined using *t*-test.

Table 5. Vitamin contents of organic and genetically modified soybeans

Vitamin (mg/100 g)	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
Vitamin B1	0.76 ± 0.06	0.71~0.82	0.83 ± 0.06	0.77~0.89	0.25	0.075~1.679
Vitamin B2	0.28 ± 0.01	0.27~0.29	0.28 ± 0.01	0.28~0.29	1.00	0.17~1.087
Folic acid	0.37 ± 0.03	336.7~396.3	0.39 ± 0.03	360.2~415.1	0.47	0.124~2.22
Vitamin E	2.82 ± 0.25	2.54~3.02	3.30 ± 0.44	2.8~3.64	0.18	0.193~12.738
Vitamin K1 (µg/100g)	16.72 ± 0.98	15.61~17.47	24.45 ± 0.39	24.18~24.9	0.0002*	6.5~207

Data are mean ± SD from three separate experiments. Literature range is from ILSI Crop Composition Database v7.0, 2019. * The significant difference (p<0.05) is examined using t-test.

Table 6. Mineral contents of organic and genetically modified soybeans

Mineral (mg/100 g)	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
Calcium	160.00 ± 13.86	146.5~174.2	223.7 ± 21.6	200.1~242.5	0.01*	116.55~488
Phosphorus	480.67 ± 41.43	432.9~506.9	467.97 ± 10.8	455.5~474.4	0.63	277~935.24
Magnesium	196.33 ± 4.91	193.5~202	199.9 ± 8.81	193.3~209.9	0.57	121~398
Potassium	1937.0 ± 24.06	1912~1960	1966.0 ± 35.03	1928~1997	0.30	833~2707.4
Iron	6.30 ± 0.30	5.95~6.48	6.53 ± 0.30	6.19~6.75	0.40	4.73~378

Data are mean ± SD from three separate experiments. Literature range is from ILSI Crop Composition Database v7.0, 2019. * The significant difference (p<0.05) is examined using t-test.

3.5. Vitamin and Mineral Contents

Five vitamins of the 2016OS and the 2016GM soybean samples were analyzed, including vitamin B1, B2, E, K1 and folic acid. The vitamin K1 content was significantly higher in the 2016GM soybean sample (24.45 ± 0.39 µg/100g) comparing to the 2016OS soybean sample (16.72 ± 0.98 g/100 µg), but these contents were within the literature range of 6.5~207 µg/100 g (Table 5). The concentrations of calcium, phosphorus, magnesium, potassium, and iron were analyzed in the 2016OS and the 2016GM soybean samples. The calcium content was significantly higher in the 2016GM soybean sample (223.7 ± 21.6 mg/100 g) comparing to the 2016OS soybean sample (160.00 ± 13.86 mg/100 g), but these contents were within the literature range of 116.55~488 mg/100 g (Table 6).

3.6. Isoflavone Contents

Isoflavones are produced via a branch of the general phenylpropanoid pathway that produces flavonoid compounds in higher plants. Soybeans are the most common source of isoflavones in human food, and the major isoflavones in soybean are genistein and daidzin. The concentrations of the total, free and modified forms of the isoflavones daidzin, genistin and glycitein were determined using high-performance liquid chromatography in the 2016OS and the 2016GM soybean samples (Table 7). There was no significant difference observed at the 5% confidence level.

3.7. Oligosaccharide and Anti-nutritional Factor Contents

The contents of stachyose and raffinose in the 2016OS and the 2016GM soybean samples were analyzed. There was no significant difference at the 5% confidence level (Table 8). We also measured the contents of phytic acid, trypsin inhibitor, and lectin in the 2016OS and the 2016GM soybean samples. There was no significant difference at the 5% confidence level between these two kinds of soybean samples (Table 8).

Table 7. Isoflavone contents of organic and genetically modified soybeans

Isoflavone (mg/100 g)	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
Daidzin	39.98 ± 9.96	28.49~46.23	29.86 ± 4.52	24.68~32.94	0.18	-
Malonyldaidzin	119.09 ± 21.83	94.21~135.05	100.49 ± 16.97	81.59~114.42	0.31	-
Acetyldaidzin	0.49 ± 0.06	0.42~0.52	0.29 ± 0.11	0.17~0.38	0.05	-
Daidzein	3.53 ± 1.50	1.81~4.54	3.07 ± 1.63	1.86~4.92	0.74	-
Total daidzein	163.09 ± 33.20	124.93~185.33	133.71 ± 12.09	119.76~141.13	0.22	6.00~306.12
Genistin	45.73 ± 9.98	34.78~54.32	29.97 ± 5.09	24.11~33.37	0.07	-
Malonylgenistin	139.25 ± 14.78	122.40~149.99	115.81 ± 23.65	88.71~132.25	0.22	-
Acetylgenistin	0.56 ± 0.17	0.38~0.71	0.37 ± 0.10	0.29~0.48	0.17	-
Genistein	3.50 ± 1.43	1.92~4.72	2.79 ± 1.35	1.87~4.34	0.57	-
Total genistein	189.04 ± 26.28	159.48~209.74	148.93 ± 20.13	125.8~162.48	0.10	3.57~283.72
Glycitin	4.62 ± 0.74	3.76~5.06	4.61 ± 1.60	2.81~5.86	0.99	-
Malonylglycitin	10.33 ± 2.18	7.98~12.28	11.66 ± 4.57	6.89~16.01	0.67	-
Glycitein	0.16 ± 0.05	0.12~0.22	0.25 ± 0.14	0.15~0.41	0.37	-
Total glycitein	15.11 ± 2.88	11.89~17.43	16.52 ± 5.97	9.85~21.35	0.73	1.41~163
Total isoflavone	367.24 ± 56.84	301.84~404.76	299.16 ± 30.81	263.92~321.00	0.14	44.73~397.21

Data were mean ± SD of three separate experiments. Literature range is from ILSI Crop Composition Database v7.0, 2019.

Table 8. Anti-nutrient contents of organic and genetically modified soybeans

Anti-Nutrient	2016OS Soybean		2016GM Soybean		*p*-Value	Literature Range
	Mean ± SD	Range	Mean ± SD	Range		
Stachyose (g/100 g)	5.87 ± 0.51	5.32~6.34	5.70 ± 0.31	5.51~6.06	0.66	0.6183~6.89
Raffinose (g/100 g)	0.91 ± 0.05	0.87~0.96	1.12 ± 0.24	0.89~1.37	0.20	0.1778~1.8542
Phytic acid (g/100 g)	1.13 ± 0 .12	0.94~1.27	1.12 ± 0.01	1.07~1.16	0.93	0.2855~2.6775
Trypsin inhibitors (TIU/mg)	77.05 ± 6.07	70.08~81.18	73.71 ± 4.45	69.88~78.58	0.48	3.23~118.68
Lectin (μg/g)	34.03 ± 6.67	26.5~39.2	43.03 ± 5.85	37.2~48.9	0.15	12.9~3600†

Data were mean ± SD of three separate experiments. Literature range is from ILSI Crop Composition Database v7.0, 2019. †The literature range of lectin content of soybean is from [17].

3.8. Pesticide Residues Present in Genetically Modified Samples

The 2016GM soybean sample contained the glyphosate and/or the glufosinate herbicide-tolerant genes (Figure 1). In this study, we examined pesticide residues in soybean samples. A total of 314 pesticide residues were examined for the 2016OS and 2016GM soybean samples. Only a low level of glyphosate residue was detected in the 2016GM soybean samples (0.08~2.12 ppm) with an average of 0.61 ppm (data not shown). However, the residue levels were well below the threshold (10 ppm) prescribed by the government provisions.

4. Discussions

To identify potential concerns for a nutrition or safety issue between genetically modified and organic soybeans for consumers in Taiwan, the compositions of the genetically modified soybean samples (2016GM) and the organic soybean samples (2016OS) purchased from Taiwan local markets in 2016 were examined and compared. Composition analysis is one of the major items for the genetically modified soybean products in Taiwan market. We analyzed the contents of proximate, amino acids, fatty acids, vitamins, minerals, isoflavones, and anti-nutritional factors between the organic soybean samples 2016OS and the genetically modified soybean samples 2016GM both purchased from Taiwan local markets in 2016. The contents of cystine and α- linoleic acid were significantly lower in the 2016GM soybean samples, and the contents of crude fat, palmitic acid, stearic acid, oleic acid, linoleic acid, vitamin K1, and calcium were significantly higher in the 2016GM soybean samples (in this study). We also analyzed the compositions of the non-genetically modified soybean samples 104-TNGM and the genetically modified soybean samples 104-GM purchased from Taiwan local markets in 2015 [18]. The contents of ash, crude protein, amino acids, myristic acid, behenic acid, phosphorus, iron and phytic acid were significantly lower in the 104-GM soybean samples, and the contents of crude fat, margaric acid, and stearic acid were significantly higher in the 104-GM soybean samples [18]. We purchased the genetically modified soybeans for two consecutive years (2015 and 2016) for composition analysis. It is interesting to note that there were eleven and nine components showed significant difference in composition contents in 2015 and 2016, respectively. However, only the content of crude fat was significantly higher for two consecutive years.

The cystine content of genetically modified soybean sample (0.31 ± 0.01 g/100 g) was significant lower than that of organic soybean sample (0.36 ± 0.00 g/100 g) (Table 3), and the content values of genetically modified soybean samples were out of the range shown in the ILSI Crop Composition Database (0.32~0.93 g/100 g). It is known that there is wide variation in protein as well as cystine contents of different soybean varieties. The cystine content of 11 soybean varieties is 0.213 ~ 0.553 g/100 g, with an average value of 0.371 g/100 g [16]. The cystine contents of Virginia, Harbinsoy, Ohio 13-177, and Peking varieties, with cystine content each of 0.213, 0.256, 0.266, 0.276 g /100 g, are outside the range of the ILSI Crop Composition Database. The results suggested that the cystine content in soybean seeds is very broad and varies greatly depending on the variety.

The composition analysis for genetically modified crops is conducted to identify potential concerns of nutritional or safety issues. When there is a statistically significant difference in the compositional analysis, the components of the differences should be evaluated based on the natural range of variation to determine its biological significance [19]. Numerous studies have also pointed out that the compositions of soybean vary significantly with different varieties and under different natural growing conditions. The nutritional profile of soybean is related to varieties. Concentrations of protein and oil in soybean seed are dependent upon its genomes [20]. There are significant difference in protein content, isoflavone content, and trypsin inhibitory activity between conventional soybeans and organic soybeans even from the same cultivar (BR258), crop season and geographic region [21]. Compared to genetically modified soybean, different agricultural practices such as conventional chemical cultivation regime and unmodified soybean cultivated using an organic cultivation regime may result in a markedly different nutritional composition of soybeans [22]. Compared to chemical fertilizer on soybean field test, compost and organic fertilizer, such as poultry and cow dung manure, showed less yield but with similar fatty acid pattern and with less insect infestation [23]. The genetic variability of the conventional breeding practice by backcross contributes the compositional difference, even though with high genetic similarity [24]. Under the different edaphoclimatic conditions, there are major differences observed between two cultivation locations. The conventional soybeans showed significantly higher protein contents and lower lipid contents, and the genetically modified soybeans showed significantly higher isoflavone contents [25]. Although significant differences in composition were found between the 2016GM and the

2016OS soybean samples, the contents were all within the range of reference values. These data suggested that the 2016GM soybeans are substantially equivalent to the 2016OS soybeans in compositions.

Glyphosate is a herbicide applied to the leaves of plants to kill both broadleaf plants and grasses. It has been found in glyphosate-resistant soybeans. A low level of glyphosate residues (0.08~2.12 ppm) was detected in the 2016GM soybean samples with an average of 0.61 ppm. The threshold of the glyphosate residue levels prescribed by the Taiwan government provisions is 10 ppm [26]. The current maximum residue levels (MRL) of glyphosate is 20 ppm in EU [27] and Japan [28]. According to the report from FAO/WHO, administration of glyphosate and its formulation products at doses as high as 2000 mg/kg body weight by the oral route, the route most relevant to human dietary exposure, was not associated with genotoxic effects in an overwhelming majority of studies conducted in mammals, a model considered to be appropriate for assessing genotoxic risks to humans [29]. The low level of glyphosate residues in the genetically modified soybeans is of no safety concern.

Genetically modified foods need to be registered and approved as admissible food by the Taiwan Food and Drug Administration to fulfill the requirement of the Act Governing Food Safety and Sanitation. Up to the end of Sep. 2019, a total of 149 genetically modified foods got approval in Taiwan, covering 5 genetically modified crops including soybean [6]. However, the attitudes of consumers in Taiwan are not friendly for genetically modified foods. Most Taiwan consumers want to buy organic foods instead of genetically modified foods. Under the new GMO labeling restrictions applied on December 31, 2015, all packaged foods, food additives, and bulk foods containing genetically modified food ingredients require mandatory labeling. Non-genetically modified food ingredients unintentionally containing more than 3% of genetically modified food ingredients are viewed as genetically modified food ingredients and must be labeled [30].

A total of 64 compositions and 314 pesticide residues of the genetically modified and the organic soybean samples was analyzed and compared. Most contents of the key components of the genetically modified soybeans had no significant difference compared with those of the organic soybeans. However, the contents of two components were significantly lower, and the contents of seven components were significantly higher in the genetically modified soybean samples. But all were within the range of reference values. Only glyphosate residue was detected in genetically modified soybean samples, but it is well below the threshold of 10 ppm.

5. Conclusion

In this study, we analyzed and compared the compositions and pesticide residues of imported genetically modified and organic soybeans purchased from Taiwan market. Less than 10 contents of key components were found to be significantly different between the two types of samples. However, they were all within the reference range. Although glyphosate residue was detected in genetically modified soybeans, it is also well below the threshold prescribed by the government. The imported genetically modified and organic soybean samples purchased from Taiwan market were substantially equivalent in compositions.

Conflicts of Interest

The authors declare no conflicts of interest.

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