

# Isoflavone and Mineral Content in Conventional and Transgenic Soybean Cultivars

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# Abstract

The objective of this study was to evaluate the differences in composition among six brands of conventional soybean and six genetically modified cultivars (GM). We focused on the isoflavones profile and mineral content questioning the substantial equivalence between conventional and GM organisms. The statement of compliance label for conventional grains was verified for the presence of genetic modified genes by real time polymerase chain reaction (PCR). We did not detect the presence of the 35S promoter in commercial samples, indicating the absence of transgene insertion. For mineral analysis, we used the method of inductively coupled plasma-optical emission spectrometry (ICP-OES). Isoflavones quantification was performed by high performance liquid chromatography (HPLC). The results showed no statistical difference between the conventional and transgenic soybean groups concerning isoflavone content and mineral composition. The concentration of potassium, the main mineral component of soy, was the highest in conventional soybeans compared to that in GM soy, while GM samples presented the highest concentration.

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### tions of iron.

### **Keywords**

Glycine max, Genetically Modified Organism, Bioactive Compounds, ICP-OES, HPLC

# 1. Introduction

Soybean (*Glycine max* Merrill), an important oilseed in different human and animal nutrition products, has high economic value in the domestic and international market. In 2012/2013, the world soybean production was 285.89 million tons. The United States, the largest grower, produced 93.08 million tons of soybeans on a cultivated area of 31.13 million hectares, and Brazil was the second largest producer, with an output of 85 million tons and occupancy of 28.25 million hectares [1].

A genetically modified organism (GMO) or transgenic organism is defined as an "organism whose genetic material has been altered by any genetic engineering technique" [2]. Genetic modification allows the production of plants with attributes of economic interest, such as resistance to biotic (e.g., pathogens, insect pests, and weeds) and abiotic stresses (e.g., salinity, drought, cold, flood, and heat); improvement of the quality of agricultural products (e.g., shelf life or prolonged storage and specific or biofortified nutritional content ); fast-growing plants; plants with greater potential for use in biofuel production; plant protection products; drug bioreactors; and/or industrial products [3].

To regulate the right to information on GM foods and considering that food labels are an important vehicle for communication between producers and society, it is important that the labeling of GMO foods complies with current legislation. In Brazil, the Decree Law No. 4680 of April 4, 2003, Ordinance No. 2658, of December 22, 2003, and Article 40 of Law No. 11.105/2005 address this issue, and regulate the mandatory labeling of foods and food ingredients for human or animal consumption produced from GMOs [2] [4] [5].

In addition to its economic and nutritional importance, soy has been studied for the presence of bioactive compounds, especially isoflavones, compounds that have a potential effect on health, reducing the risk of neuro degenerative and chronic diseases associated with aging, such as osteoporosis, cognitive dysfunction, hypertension, coronary heart disease, cancer, and menopause-related symptoms [6]. These effects have been attributed to the antioxidant and anti-estrogenic/estrogenic activity of isoflavones.

Isoflavones are a sub-class of flavonoids, which are found naturally as polyphenols and have three benzene rings in their structures. Twelve isoflavones are found in soy, comprising the aglycones daidzein, glycitein, and genistein, the glycosides daidzin, glycitin, and genistin, and their conjugated forms malonyl glucosides and acetyl glucosides [6].

Some studies have indicated that genetic variation affects the content of isoflavones in soybean cultivars and different concentrations of isoflavones can be found in different crops and different cultivation sites [7]-[11].

Nevertheless, studies on the quantification of isoflavones and mineral contents in different cultivars of genetically modified grains are scarce. Accordingly, we sought to evaluate the profile of isoflavones and minerals in conventional soybeans traded in Belo Horizonte/MG and transgenic cultivars planted in the state of Minas Gerais in the 2010/2011 season, and to correlate this profile with the presence or absence of grain genetic modification. We also assessed the conformity of the labels of the marketed soybeans samples with the legislation on mandatory GMO foods labeling.

## 2. Materials and Methods

We acquired six samples of soybeans from different suppliers in the municipality of Belo Horizonte /MG during November 2011 (SC1, SC2, SC3, SC4, SC5, and SC6). Another six samples of transgenic cultivars were provided by COPAMIL (AGRICULTURAL COOPERATIVE Mista Iraí Ltda). The following varieties were studied: Favorita-S2, Valiosa-S2, 850-S2, 811-S2, 750-C1, and 740-C1 (ST1, ST2, ST3, ST4, ST5, and ST6, respectively).

The evaluation of the labeling was performed taking into account the compliance with current legislation for food and food ingredients for human consumption or feed containing, or produced from, GMOs [2] [4] [5].

The presence of the transgene was initially verified by the detection of the 35S promoter and, when present, detection of the Roundup Ready (RR) transgene, according to ISO 21570:2005 [12]. The quantitation limit (LOD) was 0.1%.

For mineral analysis, the methodology used was that recommended by the Instituto Adolfo Lutz [13]. The minerals quantified were the following: K (potassium), Ca (calcium), P (phosphorus), Mg (magnesium), Na (sodium), Cu (Copper), Fe (iron), Mn (manganese), Zn (zinc), Cd (cadmium), Pb (lead), Ni (nickel), Ba (barium), and Cr (chromium). A standard curve was obtained by using standard solutions of these minerals at a concentration of 1000 mg/L (Merck) prepared in aqueous solution acidified at 10% (v/v). The soybeans were ground, weighed, and placed in an oven with air renewal and circulation (Marconi, mod. MA035) at 70°C, and after 12 h they were calcinated in an oven (Vulcan, mod. 3 - 1750) using a heating ramp up to 530°C. After cooling the samples, 1.0 mL of concentrated HNO<sub>3</sub> was added. This mixture was heated on a hot plate at 100°C to dryness and placed again in an oven at 375°C for 1 h. HCl (10 mL) was added to the ash obtained and gauged in a 100-mL volumetric flask with water purified with a Milli-Q system.

The samples were analyzed directly by inductively coupled plasma-optical emission spectrometry (ICP-OES; Perkin Elmer, mod. Optima 2000 DV-sampler, mod. As90plus) in Axial configuration, at 1400 kW radiofrequency power, and 0.60 L·min<sup>-1</sup> gas flow. For analysis of isoflavones, the samples were crushed in a micromill and degreased with n-hexane (HPLC grade). The extraction of isoflavones was performed according to the method proposed by Carrão-Panizzi [8] and colleagues for the extraction of isoflavones in soybeans. The separation and quantification of isoflavones were performed according to the changes in the method proposed by Berhow [14], by using a liquid chromatograph (Waters model 2690) equipped with W 600 model pump, and an automatic sample injector (model W 717 plus). An octadecyl-silica (ODS)-type reverse phase C18 column (YMC-Pack ODS AM column; 250 mm length  $\times$  0.4 mm internal diameter, particle size 5  $\mu$ m) was used for this purpose. For the separation of isoflavones we adopted a binary linear gradient system, using the following as the mobile phases: 1) methanol containing 0.025% trifluoroacetic acid (TFA) (solvent A) and 2) ultrapure deionized distilled water containing 0.025% TFA (solvent B). The initial condition of the gradient was 20% solvent A, which reached 100% after 40 min, then reduced to 20% after 41 min, and remained in this condition up to 60 min. Therefore, the total time of analysis for each sample was 60 min. The mobile phase flow rate was 1.0  $mL \cdot min^{-1}$  and the temperature throughout the analysis was maintained at 25°C. The detection of isoflavones was performed using a photo diode array detector (Waters model W 996) adjusted to the wavelength of 260 nm. To identify the peaks corresponding to each of the twelve different forms of isoflavones, the following standards were used: daidzin, daidzein, genistin, genistein, glycitin, glycitein, and also standards of their acetyl and malonyl conjugates, (Sigma and Fuji) solubilized in methanol (HPLC grade) at the following concentrations: 0.00625 mg/mL, 0.0125 mg/mL, 0.0250 mg/mL, 0.0500 mg/mL, and 0.1000 mg/mL. To quantify the 12 types of isoflavones by external standardization (peak area), standards were used as reference. The identification of the peaks corresponding to each of the twelve forms of isoflavones in the samples was performed by comparison with the spectra and retention times of the standards.

Statistical analysis was performed using analysis if variance (ANOVA) and Tukey's test for comparison of the means, as well as the Statistica 7.0 program (STATSOFT, 2004), with a significance level of 5%.

## 3. Results and Discussion

Article 40 of Law No. 11.105/2005 defines that "food and food ingredients for human consumption or animal feed containing or produced from GMOs or derivatives should contain information on their labels accordingly" [4]. Decree 4680/2003 regulates the mandatory labeling of foods and food ingredients for human consumption or animal feed containing or produced from GMOs that contain GMOs above the limit of 1% of the product. These products should bear on their labels prominently the words: "(name of product) transgenic", "contains (name of ingredient or ingredients) transgenic (s)", or "product produced from (name of product) transgenic" as well as the symbol for GM, which consists in a yellow triangle with the letter "T" inside, defined by Ordinance No. 2658, December 22, 2003 [4] [5].

Initially, we observed that the label statements in the commercial soybean samples did not comply with the law. However, the results of the genetic analysis did not show the presence of the 35S promoter. Therefore, there was no need to investigate RR gene alterations, as described in the methodology. Thus, since the tested samples effectively contained no detectable amounts of GMOs, the brands sampled in local suppliers were in accordance with the law.

It is interesting to note that although 76% of soybeans grown in Brazil are from transgenic cultivars, the conventional product continues to be prominent in local trades. In a study by Branquinho and colleagues [15], it was shown that 28.3% of the samples tested contained transgenic soybean [(n = 68) in 240 foods derived from soybeans analyzed between 2004 and 2007]. Quantitative analysis revealed GMO content between 0.05% and 1% in 43 (63.2%) samples, and more than 1% in 25 (36.8%) samples. There was no indication on the label of the presence of GM material and; therefore, the labels were not in accordance with the law. These results differ from ours probably because the authors analyzed products derived from soy (type 1 products), and not soybeans, which were the object of our study.

Over the last decade, other studies have assessed the presence of GMOs in food available in the consumer market. Cardarelli *et al.* [16] analyzed 89 food products that contained soy and/or corn ingredients in samples originating from different cities in Brazil. The presence of soybean RR was found in 16 samples such as soy extract, pastes, sauces, dehydrated soups, and uncooked soybean feed. Of the total number of samples analyzed by Cardarelli and colleagues [16], 15 were raw soybeans and only one was positive for the presence of the 35S promoter. The results and methodology of this study are similar to those of the present study. Subsequently, the positive sample found by Cardarelli *et al.* [16] was evaluated for the presence of the RR transgene, which was then confirmed.

Brod and colleagues [17] have produced a series of articles evaluating the presence of GM soy in different food products marketed in Florianópolis, Brazil. In 2007, they analyzed 37 samples of soy products including six samples of flour, six samples of infant formula, and 25 samples of soymilk powder. The results were positive for the presence of RR soybean in four samples of defatted soy flour and 15 samples of soymilk powder [18].

In 2007, Brod and Arisi [18] analyzed 32 meat additives containing soy proteins. Twenty-five were positive for lecithin, confirming the presence of soy in the amplified DNA, and 15 of these were positive for RR, confirming the presence of GM soy.

In 2008, Brod and Arisi [19] analyzed 62 samples of soy isolates, of which 37 were of textured soy protein (TSP) and 25 were of soybean extract powder. Forty samples were positive for RR but only two contained more than 1% RR soybean.

It is interesting to note that these studies were conducted primarily with soy products, in which the presence of GM soy lecithin and RR was detected but not the 35S promoter. These results suggest that the use of GM soybeans is directed to processed foods.

Regarding the glycoside daidzin (SC 4.61  $\pm$  1.88 and ST 6.76  $\pm$  3.74 mg·100 g<sup>-1</sup> of defatted sample) and its corresponding aglycone form daidzein (SC 55.40  $\pm$  12.64 and ST 67.14  $\pm$  31.95 mg·100 g<sup>-1</sup> of defatted sample), there was no statistically significant difference (p > 0.05) when comparing the ST (GM soy) and SC (commercial soybean) sample groups, which is in accordance with the results of Zhou and colleagues [11], who also found no significant difference between samples of transgenic soybeans and conventional soybeans for these forms of isoflavones. There were detected all forms of isoflavones in this study; therefore, the three forms of isoflavones and aglycone glycitein were not quantified. Because the study focused on unprocessed soybeans, the finding of acetyl conjugates was expected, as they are only present in processed soy products.

The work of Zhou and colleagues [11] demonstrated, by a mixed model, regional differences in isoflavone content; all levels of isoflavones were higher in southern Brazil when compared with the levels found in the samples from northern Brazil. This model attributed the variation in the composition of the isoflavones mostly to a combination of parameters such as region, year, and phenotype, which were statistically different (p < 0.05) for all the isoflavones evaluated. These data confirm that multiple factors are associated with the variation in the content of isoflavones, including environmental factors such as local culture and especially the temperature during grain filling; genetic factors inherent to the soybean cultivars also influence the accumulation of isoflavones in the grains. This has been consistently observed in recent studies [7]-[10], establishing that the formation and accumulation of isoflavones is affected by many biotic and abiotic factors.

The absence of statistically significant difference (p > 0.05) between the samples of GM and conventional soy samples found in this study for these two isoflavones (daidzin and daidzein) shows the absence of any effect attributed to the transgene.

In contrast, when we compared the levels of other isoflavones in samples of transgenic cultivars and conventional commercial soy we observed a significant difference (p < 0.05), despite the large intragroup variability observed (**Table 1**). For malonyl malonyl-glycitein forms (ST 21.91  $\pm$  2.68 and SC 15.95  $\pm$  8.91 mg·100 g<sup>-1</sup>), malonyl-daidzein (ST 167.06  $\pm$  28.25 and SC 81.40  $\pm$  50.78 mg·100 g<sup>-1</sup>), and malonyl-genistein (ST 206.16  $\pm$  25.43 and SC 113.32  $\pm$  46.22 mg·100 g<sup>-1</sup>), we observed higher average levels in transgenic cultivars than in conventional commercial samples.

However, for both glycosides glycitin (ST 7.53  $\pm$  1.99 and SC 11.43  $\pm$  4.74 mg·100 g<sup>-1</sup>) and genistein (ST 36.97  $\pm$  7.77 and SC 58.08  $\pm$  27.28 mg·100 g<sup>-1</sup>), the levels observed were statistically higher for the transgenic cultivars. The mean levels of genistein observed were 2.95  $\pm$  1.24 mg·100 g<sup>-1</sup> in transgenic cultivars, and 4.83  $\pm$  2.41 mg·100 g<sup>-1</sup> for conventional commercial cultivars, which indicated that these, on average, had higher levels of this bioactive isoflavone. However, the wide range of contents of isoflavones in soybeans and transgenic cultivars, especially in conventional commercial cultivars, as verified by the intragroup standard deviation, may have interfered with this significance. The variation in the concentration of isoflavones, especially in the conventional commercial by the potential heterogeneity both with regard to genotype and the place and time of planting, growing conditions, and even storage conditions.

The profile of total isoflavones for ST cultivars and SC cultivars (ST 502.60  $\pm$  65.63 and SC 358.90  $\pm$  135.21 mg·100 g<sup>-1</sup>) was similar to that observed by Bhom *et al.* (2008) (ST 211.2 and SC 182.2 mg·100 g<sup>-1</sup>), who found that, even though lower levels of these compounds were observed in GM varieties, the total isoflavone content was higher. It is important to note that, in our study, this difference was statistically significant. In another study by Barbosa *et al.* [20], the authors found 381  $\pm$  3 mg 100 g<sup>-1</sup> of total isoflavones in defatted soybean flour obtained from conventional commercial soybeans, which is similar to our results for conventional commercial soybean, because isoflavones were assessed on degreased samples. Duke *et al.* [21] performed a study using a different approach to evaluate the response of transgenic cultivars DP 5806 RR and Asgrow 3701 RR subjected to different environmental conditions but similar herbicide applications. The authors observed a wide variation in the levels of isoflavones between the two cultivars, but not within the same cultivar with different herbicide applications, which suggests that this wide range of variation among cultivars is independent of the presence of the transgene.

| Samples | Daidzein                 | Genistein                | Total Isoflavones              |
|---------|--------------------------|--------------------------|--------------------------------|
|         |                          | mg/100g                  |                                |
| ST1     | $6.53\pm0.27^{\rm c}$    | $4.34\pm0.14^{\rm d}$    | $591.29 \pm 14.44^{\rm a}$     |
| ST2     | $2.32\pm0.00^{\text{g}}$ | $1.13\pm0.02^{\rm h}$    | $533.73 \pm 13.56^{b,c}$       |
| ST3     | $5.58\pm0.19^{\rm d}$    | $3.22\pm0.01^{\text{e}}$ | $549.08 \pm 3.55^{b}$          |
| ST4     | $2.17\pm0.02^{\rm g}$    | $1.64\pm0.01^{\rm g}$    | $413.86 \pm 7.55^{\rm f,g}$    |
| ST5     | $6.51\pm0.01^{\rm c}$    | $4.08 \pm 0.08^{\rm d}$  | $491.94\pm0.58^{\rm d}$        |
| ST6     | $4.58\pm0.01^{\text{e}}$ | $3.28\pm0.05^{\text{e}}$ | $435.72 \pm 1.59^{\text{e,f}}$ |
| SC1     | $5.30\pm0.07^{\rm d}$    | $4.28\pm0.04^{\rm d}$    | $227.22\pm0.39^{h}$            |
| SC2     | $3.36\pm0.06^{\rm f}$    | $2.07\pm0.04^{\rm f}$    | $508.09 \pm 4.43^{\rm c,d}$    |
| SC3     | $11.16\pm0.18^{\rm a}$   | $7.93\pm0.02^{\rm a}$    | $447.63 \pm 1.19^{\circ}$      |
| SC4     | $9.47\pm0.01^{\text{b}}$ | $6.05\pm0.13^{\rm c}$    | $434.05 \pm 1.90^{e,f}$        |
| SC5     | $9.74\pm0.01^{\text{b}}$ | $6.80\pm0.08^{\rm b}$    | $392.64 \pm 3.40^{g}$          |
| SC6     | $1.52\pm0.07^{\rm h}$    | $1.83\pm0.09^{\rm f,g}$  | $143.78\pm0.77^{\rm i}$        |
| ALL     | $5.69\pm3.09$            | $3.89\pm2.10$            | $430.75 \pm 127.24$            |

**Table 1.** Concentrations of aglycones (daidzein and genistein) and total isoflavones (mg 100  $g^{-1}$  defatted samples) in transgenic and conventional commercial soybean cultivars.

Means followed by the same lower case letter in columns do not differ according to the Tukey's test at 5% significance. The results are expressed in  $mg \cdot 100 g^{-1}$  of dry, defatted sample. The GM soybeans ST1-ST6 correspond to the following cultivars, respectively: BRS Favorita, BRS Valiosa, BRSMG 850G, BRSMG 811C, BRSMG 750S, and BRSMG 740S. ST = Transgenic; SC = Conventional.

When grouping isoflavones by their radical groups, the average percentages observed for the different cultivars (19.82 and 38.61 mg·100 g<sup>-1</sup> of  $\beta$ -glycosides, 78.68 and 58.11 mg·100 g<sup>-1</sup> of malonylglycosides, 1.49 and 3.28 mg·100 g<sup>-1</sup> of aglycones and no acetylglycosides, for transgenic soybean cultivars and conventional commercial cultivars, respectively) were in agreement with the findings of Carrão-Panizzi [9] for raw grains when the values were converted into percentage of total isoflavones (19.15, 79.26, and 1.55 mg·100 g<sup>-1</sup>). However, Barbosa *et al.* [20] found different proportions of total isoflavones: 42.8 and 39.1 mg·100 g<sup>-1</sup> of  $\beta$ -glycosides, 52.5 and 50.6 mg·100 g<sup>-1</sup> of malonylglycosides, 4.0 and 7.2 mg·100 g<sup>-1</sup> of aglycones, and 1.0 and 2.9 mg·100 g<sup>-1</sup> of acetylglycosides in soybeans and defatted flour, respectively. However, these authors did not have information about the presence of transgenes in the tested samples. Bavia *et al.* [22] using the same methodology, found different values for different cultivars and variation in the proportion in raw grains:  $\beta$ -glycosides from 37.9% to 47.8%, malonylglycosides from 40.8% to 48.1%, and aglycones from 11.4% to 16.7%; total isoflavones from 89.63% to 200.24 mg·110 g<sup>-1</sup> of dry matter.

Regarding the aglycones, the values found in this study were similar to those obtained by Benedetti [23] in defatted soybeans, which represented 1.8% of total isoflavone content, and to those found by Wang & Murphy [24], in which the aglycone levels were between 1% and 3% of total isoflavones. The average total aglycone was 9.6 mg·100 g<sup>-1</sup>. This result was similar to that found by Silva *et al.* [25], which was 9.19 mg·100 g<sup>-1</sup>, and higher than that reported by Carrão-Panizzi *et al.* [8], which was 4.0 mg·100g<sup>-1</sup>. These variations are explained by the cultivar studied and the environmental cultivation conditions.

The results of the analysis of micro- and macrominerals are summarized in **Table 2** and **Table 3**, respectively. When comparing the results of the analysis of macro- and micro-minerals, with the samples divided into conventional and transgenic cultivars, it was possible to observe significant differences between groups for the following minerals: potassium (p < 0.003), chromium (p < 0.007), and iron (p < 0.013). The minerals Pb, Ni, and Cd were not detected or were classified as trace elements.

| Mineral          | Calcium                           | Magnesium                    | Phosphorus                          | Sodium                        | Potassium                      |
|------------------|-----------------------------------|------------------------------|-------------------------------------|-------------------------------|--------------------------------|
| Samples          |                                   |                              | mg/Kg                               |                               |                                |
| Favorita-S2 (T)  | $1606.12\pm87.38^{b,c}$           | $1789.75 \pm 127.48^{b}$     | $4728.30 \pm 153.29^{c,d}$          | $100.12\pm7.74^{a,b}$         | $14281.81 \pm 1917.94^a$       |
| Valiosa-S2 (T)   | $1419.50 \pm 43.96^{\text{b,c}}$  | $2049.40 \pm 20.84^{a,b}$    | $5698.98 \pm 31.23^{a,b}$           | $131.53 \pm 13.65^{a,b}$      | $12119.69\pm756.75^{a}$        |
| 850-S2 (T)       | $1655.84 \pm 591.21^{\text{b,c}}$ | $1689.52 \pm 571.29^{b}$     | $3736.44 \pm 60.59^{e}$             | $113.54 \pm 15.95^{a,b}$      | $12280.15\pm 65.39^{a}$        |
| 811-S2 (T)       | $1101.81 \pm 446.06^{\rm c}$      | $1730.69 \pm 610.22^{b}$     | $5718.49 \pm 273.91^{a.b}$          | $121.72 \pm 27.63^{a,b}$      | $13544.75 \pm 1144.75^{a}$     |
| 750-C1 (T)       | $2167.74 \pm 259.42^{a,b}$        | $2854.71 \pm 326.47^{a}$     | $5252.64 \pm 105.52^{b,c}$          | $121.06\pm7.02^{a,b}$         | $12307.43 \pm 1127.58^{a}$     |
| 740-C1 (T)       | $2908.53 \pm 52.14^{\rm a}$       | $2464.76 \pm 67.56^{a,b}$    | $4980.12 \pm 15.43^{\text{b,c,d}}$  | $145.51\pm5.91^{\rm a}$       | $12851.25\pm 286.19^{a}$       |
| Commercial 1 (C) | $1240.22 \pm 37.37^{\circ}$       | $2349.18 \pm 39.77^{a,b}$    | $5372.40 \pm 489.95^{b}$            | $123.65\pm3.23^{a,b}$         | $15158.29\pm 3040.44^{a}$      |
| Commercial 2 (C) | $2971.15 \pm 124.07^{a}$          | $2625.78 \pm 99.17^{a,b}$    | $4620.21 \pm 87.41^{d}$             | $148.18\pm13.36^{\mathrm{a}}$ | $13605.48 \pm 706.93^{a}$      |
| Commercial 3 (C) | $1560.34 \pm 459.13^{\text{b,c}}$ | $1900.22 \pm 510.69^{b}$     | $5978.96 \pm 125.24^{a}$            | $107.94 \pm 12.42^{a,b}$      | $12797.47\pm 352.74^{a}$       |
| Commercial 4 (C) | $1796.07 \pm 249.56^{\text{b,c}}$ | $2201.59 \pm 296.17^{a,b}$   | $5694.39 \pm 267.23^{a,b}$          | $85.39\pm35.62^{\mathrm{b}}$  | $15302.11 \pm 610.67^a$        |
| Commercial 5 (C) | $1928.78 \pm 192.67^{\text{b,c}}$ | $2288.56 \pm 166.66^{a,b}$   | $6151.77 \pm 139.01 \ ^{a}$         | $133.06\pm4.99^{a,b}$         | $15536.83 \pm 140.98^{a}$      |
| Commercial 6 (C) | $1618.48 \pm 49.27^{\text{b,c}}$  | $2279.14 \pm 36.10^{a,b}$    | $4935.65 \pm 174.28^{\text{b,c,d}}$ | $117.89 \pm 15.93^{\rm a,b}$  | $13825.31 \pm 160.56^{a}$      |
| Mean ST          | $1809.92 \pm 660.59^{\rm A}$      | $2096.47 \pm 540.80^{\rm A}$ | $5019.16 \pm 704.64^{\rm A}$        | $123.29 \pm 16.43^{\rm A}$    | $12897.52 \pm 1209.12^{\rm B}$ |
| Mean SC          | $1852.50 \pm 593.35^{\rm A}$      | $2274.08 \pm 306.96^{\rm A}$ | $5458.90 \pm 601.02^{\rm A}$        | $118.31 \pm 26.89^{\rm A}$    | $14370.91 \pm 1518.34^{\rm A}$ |
| Total Mean       | $1831.21 \pm 619.21$              | $2185.28 \pm 442.64$         | $5239.03 \pm 682.89$                | $120.80\pm22.11$              | $13634.22 \pm 1545.34$         |

| Table 2. Content of macrominerals in | n transgenic and | conventional | commercial | soybean | cultivars | grown an | nd market | ed in th | le |
|--------------------------------------|------------------|--------------|------------|---------|-----------|----------|-----------|----------|----|
| state of Minas Gerais.               |                  |              |            |         |           |          |           |          |    |

T = Transgenic; C = Conventional; ST = Grouped Transgenic Soybeans; SC = Grouped Conventional Soybeans. Mean values (±standard deviation) with the same letter do not differ significantly according to the Tukey's test at 5% significance. Lower case letters, comparison of samples by mineral. Uppercase letters, comparison between GM and commercial soybeans by mineral.

| Mineral          | Barium                      | Copper                        | Chromium                 | Iron                               | Manganese                       | Zinc                               |  |
|------------------|-----------------------------|-------------------------------|--------------------------|------------------------------------|---------------------------------|------------------------------------|--|
| Samples          |                             |                               | mg/Kg                    |                                    |                                 |                                    |  |
| Favorita-S2 (T)  | $5.93\pm0.36^{\rm c}$       | $91.19\pm4.56^{a}$            | $0.42\pm0.10^{\text{b}}$ | $101.73\pm6.48^{b}$                | $21.15\pm1.32^{a,b,c,d}$        | $98.49\pm0.24^{a}$                 |  |
| Valiosa-S2 (T)   | $4.93\pm0.17^{\rm c}$       | $13.77\pm0.31^{\text{b,c}}$   | $0.43\pm0.15^{\text{b}}$ | $74.33 \pm 0.75^{\rm f,g}$         | $21.10\pm0.03^{a,b,c,d}$        | $56.54\pm0.49^{b,c}$               |  |
| 850-S2 (T)       | $24.57\pm7.86^{\rm a}$      | $11.89 \pm 1.91^{\text{b,c}}$ | $0.61\pm0.27^{\text{b}}$ | $127.38\pm8.79^a$                  | $22.53 \pm 7.09^{a,b,c}$        | $44.96\pm6.09^{\text{d},\text{e}}$ |  |
| 811-S2 (T)       | $5.44\pm0.38^{\rm c}$       | $12.75\pm0.53^{\text{b,c}}$   | $0.53\pm0.14^{\text{b}}$ | $94.16\pm3.34^{\text{b,c,d}}$      | $12.93 \pm 4.06^{\text{d}}$     | $50.74\pm3.37^{\text{b,c,d}}$      |  |
| 750-C1 (T)       | $12.04\pm2.51^{\text{b,c}}$ | $11.59\pm1.48^{\text{b,c}}$   | $0.61\pm0.16^{\text{b}}$ | $78.6\pm1.75^{\text{e,f,g}}$       | $15.64\pm2.01^{\text{c,d}}$     | $47.13\pm7.64^{\text{c,d,e}}$      |  |
| 740-C1 (T)       | $5.51\pm0.27^{\rm c}$       | $12.18\pm0.3^{\text{b,c}}$    | $0.19\pm0.07^{b}$        | $93.52\pm1.06^{\text{b,c,d,e}}$    | $16.72\pm0.07^{\text{b,c,d}}$   | $41.93\pm3.23^{\text{d},\text{e}}$ |  |
| Commercial 1 (C) | $5.98\pm0.23^{\rm c}$       | $15.92\pm0.54^{\text{b}}$     | $0.50\pm0.17^{\text{b}}$ | $82.53 \pm 11.35^{\text{d,e,f,g}}$ | $23.16\pm0.63^{a,b,c}$          | $45.76\pm5.33^{\text{c,d,e}}$      |  |
| Commercial 2 (C) | $11.98\pm0.38^{\text{b,c}}$ | $12.63\pm0.16^{\text{b,c}}$   | $0.98\pm0.63^{\text{b}}$ | $86.19\pm1.43^{\mathrm{c,d,e,f}}$  | $29.25\pm1.1^{\text{a}}$        | $46.92 \pm 1.82^{\text{c,d,e}}$    |  |
| Commercial 3 (C) | $11.78\pm3.66^{\text{b,c}}$ | $9.48 \pm 2.57^{\rm c}$       | $0.73\pm0.39^{\text{b}}$ | $75.19\pm2.76^{\rm f,g}$           | $15.07\pm3.91^{\text{c,d}}$     | $56.22\pm1.14^{\text{b,c}}$        |  |
| Commercial 4 (C) | $12.57 \pm 2.22^{b,c}$      | $10.87\pm1.74^{\text{b,c}}$   | $0.55\pm0.11^{\text{b}}$ | $70.82\pm3.95^{\text{g}}$          | $17.71\pm2.56^{\text{b,c,d}}$   | $51.37\pm3.07^{\text{b,c,d}}$      |  |
| Commercial 5 (C) | $15.72\pm2.08^{\text{b}}$   | $11.97 \pm 1.20^{\text{b,c}}$ | $0.64\pm0.23^{\text{b}}$ | $77.66 \pm 1.27^{\rm f,g}$         | $17.91 \pm 1.92^{\text{b,c,d}}$ | $60.20 \pm 1.02^{\text{b}}$        |  |
| Commercial 6 (C) | $6.3\pm0.95^{\rm c}$        | $14.96\pm0.29^{b}$            | $1.18\pm0.43^{a}$        | $99.22\pm4.73^{\text{b,c}}$        | $24.53\pm0.15^{a,b}$            | $38.72\pm2.38^{\text{e}}$          |  |
| Mean ST          | $9.74\pm7.80^{\rm A}$       | $25.56\pm30.26^{\text{A}}$    | $0.46\pm0.20^{\text{B}}$ | $94.95\pm18.23^{\rm A}$            | $18.34\pm4.61^{\rm A}$          | $56.63\pm20.18^{\rm A}$            |  |
| Mean SC          | $10.72\pm3.96^{\rm A}$      | $12.64\pm2.57^{\rm A}$        | $0.76\pm0.40^{\rm A}$    | $81.94\pm10.49^{\text{B}}$         | $21.27\pm5.29^{\rm A}$          | $49.87\pm7.65^{\rm A}$             |  |
| Total Mean       | $10.23\pm6.12$              | $19.10\pm22.16$               | $0.61\pm0.35$            | $88.44 \pm 16.08$                  | $19.81\pm5.11$                  | $53.25 \pm 15.43$                  |  |

Table 3. Content of trace elements in transgenic and conventional commercial soybean cultivars grown and marketed in the state of Minas Gerais.

T = Transgenic; C = Conventional; ST = Grouped Transgenic Soybeans; SC = Grouped Conventional Soybeans. Mean values (± standard deviation) with the same letter do not differ significantly according to the Tukey's test at 5% significance. Lower case letters, comparison of samples by mineral. Uppercase letters, comparison between GM and commercial soybeans by mineral.

In study by Zobiole *et al.* [26] found lower values for macro- and micro-nutrients analyzed in soybeans that had undergone genetic modification compared to their near-isogenic cultivars, regardless of the application of glyphosate; however, these studies evaluated the plant (shoot and root) and not the seeds. In study by Shinonaga *et al.* [27], the uptake and translocation of trace elements (Co, Se, Rb, Sr, Ru, Rh, and Cs) in maturing soybean plants cultivated on soil were studied over 360 h under diurnal conditions after the administration of a multitracer. In study by Vieira *et al.* [28] evaluated six soybean cultivars and observed that K was the most abundant mineral in all cultivars, with a maximum of 1824.02 mg·100 g<sup>-1</sup> in the Iguaçu cultivar, and a minimum of 1567.05 mg·100 g<sup>-1</sup> for the EMBRAPA-4 cultivar. The contents of P, Ca, and Mg ranged from 454.71 to 503.84 mg·100 g<sup>-1</sup>, 170.19 to 313.93 mg·100 g<sup>-1</sup> and 214.36 to 259.97 mg·100 g<sup>-1</sup>, respectively. The concentrations of Fe, Mn, and Na varied between 13.39 and 19.12 mg·100 g<sup>-1</sup>, 1.75 and 2.79 mg·100 g<sup>-1</sup>, and 11.73 and 12.08 mg·100 g<sup>-1</sup>, respectively. In this study, the high variation in mineral contents between cultivars may be related to genetic factors intrinsic to each cultivar as well as growing conditions, climate, soil, and fertilizers. We also observed that for these minerals, the values obtained in this study were lower than those found by Vieira *et al.* [28] in their samples.

# 4. Conclusions

The six brands of conventional soybean marketed in the municipality of Belo Horizonte were labeled for the presence of genetically modified organisms, in accordance with current legislation.

Although differences among groups were observed for some minerals, the same trend was observed within the group, which could be explained by genetic differences among cultivars as well as by environmental conditions during cultivation. The highest levels of potassium and the main mineral present in soy were found in conventional cultivars when compared with transgenic cultivars, which in turn had higher content of iron. The variation in total isoflavone contents of soybeans from local suppliers confirms the need for labels to bear information regarding these levels. In addition, information is needed regarding unknown parameters such as variety, cultivation region, and maturation of the commercially available grains.

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