Chemical composition and effects of micronized corn bran on iron bioavailability in rats
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Abstract
The degermination of corn grains by dry milling generates 5% of a fibrous residue. After segregation and micronization, corn bran becomes a potential source of dietary fiber consumption. However, its effect on iron bioavailability has not been reported in the literature. The objective of the present study was to determine the nutritional composition of corn bran and its effects on iron bioavailability using the hemoglobin depletion-repletion method in rats. The animals were divided into two groups: cellulose (control) and corn bran (experimental). The bran had high content of total dietary fiber, especially the insoluble fraction, and low phytate content. Hemoglobin uptake did not differ between groups at the end of repletion period, and the iron relative bioavailability value of the corn bran diet was 104% in comparison to that of the control group. The product evaluated proved to be a potential source of dietary fiber and it showed no negative effects on iron bioavailability.

Keywords: dietary fiber; phytate; mineral.

1 Introduction
Fiber-rich foods or dietary fiber (DF) can interfere in mineral bioavailability such as iron, especially in the presence of phytate due to possible formation of insoluble compounds (Schlemmer et al., 2009; Cercamondi et al., 2013). The hemicellulose fraction of dietary fiber can uptake metal ions due to formation of bonds with carboxylic groups of uronic acids and/or hydroxyl groups (Nawirska & Uklanska, 2008). However, the DF itself does not significantly influence iron absorption; the inhibitory effect of cereal bran is attributed almost totally to its high phytate content (Fantini et al., 2008).

The benefits of DF and the grain bran in human health have been extensively described (Mann & Cummings, 2009), and among them is the control of disorders associated with the metabolic syndrome. DF plays an important role in the regulation of body weight, food intake, obesity, arterial blood pressure, glucose homeostasis, insulin sensitivity, type 2 diabetes mellitus, intestinal constipation, cancer, and inflammation mediators. DF also improves serum lipid profile and hence reduces the risk factors of cardiovascular diseases (Galisteo et al., 2008). Corn is an energetic food component of human and animal diets and is considered an important DF source, especially insoluble fibers (hemicellulose, cellulose and lignin) (Paes, 2006).

During the corn dry-grind process, there is an accumulation of a portion of fibrous bran (5% of the total processed grains), which results from the grain degermination process and has not been well studied or used. This product is basically composed of the anatomical fractions of the pericarp and the tip cap of the corn kernel. A small quantity of endosperm is also present, especially the aleurone layer (Paes, 2006), where the phytate is concentrated (Mjoun et al., 2008). Considering that the pericarp is constituted of hemicellulose (67%), cellulose (23%), and lignin (0.1%) (Paes, 2006); this product can be a potential source of DF (Sayer et al., 2013). In view of the possibility of adding value to corn, the corn dry milling industries micronize this bran reducing its particles size, thus making it more attractive to be incorporated in the formulations of rich-fiber food products. Little is known, however, about its nutritional value, the phytate content, and the potential effects on mineral bioavailability. Therefore, the present study aimed to evaluate the chemical composition and the influence of micronized corn bran on iron bioavailability and its potential use as a source of dietary fiber.

2 Materials and methods
The samples of micronized corn bran were obtained from the Corn Processing Unit of the Integrated Cooperative (Andirá, PR, Brazil) during the corn dry processing and supplied by EMBRAPA Maize and Sorghum (Sete Lagoas, MG, Brazil).

2.1 Determination of centesimal composition
Moisture, protein, lipids, starch, total dietary fiber, and fractions were determined (Association of Official Analytical Chemists, 1997). Ash content was determined by sample calcination in muffle furnace at 550 °C until constant weight (Pregnolatto & Pregnolatto, 1985). The mineral content in the samples was determined (Gomes et al., 2003) by digesting 1 g of the sample in concentrated nitric acid at 130 °C for 16 hours until a clear solution was formed. The concentration of iron,
zinc, calcium, manganese, phosphorus, and copper in the corn bran was determined by atomic absorption spectrophotometry (spectrophotometer GBC 908 AA - GBC/Germany, Analytical, São Paulo, Brazil). Sodium and potassium were determined by flame spectrometry (Gomes et al., 2003). For phytate analysis, the samples were lyophilized in freeze-dryer before extraction of different inositol phosphates. The analysis was carried out according to Official Analytical Chemists (Association of Official Analytical Chemists, 1997), and the chromatographic method (ionic pair, Ultrasep ES 100 RP18, 2x250 mm) proposed by Sandberg & Ahderinne (1986).

2.2 Biological assay

Forty-eight male weaning Wistar rats (Rattus norvegicus, albinus variety, Rodentia class), weighing between 50 and 60 g, were kept in individual cages in a controlled environment at 22±2 °C and 12 h light/dark cycle. The experiment was carried out in two phases: depletion, to induce anemia, and hemoglobin repletion, to recover it. In the depletion phase (Association of Official Analytical Chemists, 1997; Enes et al., 2014), the animals fed a diet with free-iron mineral mix to induce iron-deficiency anemia, based on the AIN-93G diet (Reeves et al., 1993). Deionized water was offered ad libitum. Body weight was weekly monitored and food ingestion was controlled (pair-fed). After 21 days of depletion, hemoglobin and hematocrit were quantified, and the anemic animals were divided into 6 groups of 8, according to the hemoglobin concentration. In this phase, the animals were fed the experimental diets for 14 days, with controlled daily intake of approximately 15 g, and deionized water ad libitum. Animals’ weight and food intake were weekly monitored, to calculate weight gain and food efficiency ratio (FER= weight gain (g)/food consumption (g) x 100). The relative bioavailability value (RBV) was calculated by establishing the relationship between the angular linear coefficient obtained for the micronized corn fiber, DF source (experimental group), and the linear coefficient referring to the control diet (cellulose group) obtained from the linear regression analysis. The reference standard (ferrous sulfate in the presence of cellulose) was considered equal to 100%, and the ferrous sulfate RBV in the presence of fibrous corn bran was calculated.

At the end of this period, hemoglobin and hematocrit were determined calculating the hemoglobin gain by the difference between values obtained in the repletion and depletion phases. Weight gain and FER were also calculated. The Guide for Care and Use of Laboratory Animals was followed, and the project was approved by the Animal Experimentation Ethics Committee (CETEA) of the Federal University of Minas Gerais, (protocol number 111/2009).

2.3 Composition and preparation of experimental diets

The compositions of the control and experimental diets are shown in Table 1. The depletion diet was based on the AIN-93G diet (Reeves et al., 1993) containing iron-free mineral mix. For the repletion phase, the control diets were also based on the AIN-93G. Three diets containing graded levels of iron, 6, 12, or 24 mg Fe.kg⁻¹ diet as ferrous sulfate (FeSO₄, 7H₂O) that contained 0.2014 mg Fe.mg⁻¹ were prepared. An iron-free mineral mixture was used, and the DF source (microfine cellulose) was replaced with micronized corn bran, considering its total dietary fiber (TDF) content 73.4% (Table 2), in order to supply the same 5% DF as the control diet. Knowing the amount of bran to be included in the diets (68.0 g.kg⁻¹) and considering that the iron concentration was 27.38 mg kg⁻¹, the FeSO₄·7H₂O amount to be added so that the diets offered 6, 12, or 24 mg Fe.kg⁻¹ was calculated. Albumin was used as protein source to substitute for casein in the depletion and repletion diets, due to its lower iron content. The diets were isocaloric.

The ingredients were individually weighed in a semi-analytical balance (GEHAKA®, BG2000, São Paulo, Brazil) with

Table 1. Composition of experimental diets (g.kg⁻¹).

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Depletion Diet</th>
<th>Repletion Diet</th>
<th>Iron levels (mg.kg⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Modified AIN93-G (*)</td>
<td>Repletion Diet</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Control Diet (Cellulose)</td>
<td>Experimental Diet (Corn Bran)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>12</td>
<td>24</td>
</tr>
<tr>
<td>Albumin*</td>
<td>200</td>
<td>200</td>
<td>200</td>
</tr>
<tr>
<td>Dextrinized Starch*</td>
<td>132</td>
<td>132</td>
<td>132</td>
</tr>
<tr>
<td>Sucrose*</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Soy Oi†</td>
<td>70</td>
<td>70</td>
<td>70</td>
</tr>
<tr>
<td>Microfine Cellulose (Fiber)*</td>
<td>50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>Corn Bran*</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Iron-free Mineral Mix#</td>
<td>35</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>Vitamin mix*</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>L-Cystine*</td>
<td>3</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Choline Bitartrate*</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>Corn Starch*</td>
<td>397.5</td>
<td>397.5</td>
<td>397.5</td>
</tr>
<tr>
<td>FeSO₄·7H₂O (mg)</td>
<td>-</td>
<td>29.8</td>
<td>59.6</td>
</tr>
</tbody>
</table>

(∗) Adapted from Reeves et al., 1993; iron-free, with albumin as a substitute for casein; Brand/ supplier: 14 Maximum / ARVE -Viçosa/MG - Brazil; 15 – Açúcar União / Viçosa market; 16 – SOYA / Viçosa market; 17 – Rhoster - Indústria e Comércios Ltda; 18 – Unit of Corn Processing of the Integrated Cooperative – Andirá/PR; 19 – based on Mineral Mix AIN-93G; 20 – Pink Alimentos Belo Horizonte / Viçosa market; 21 – Vetec / Veteck – RJ.

readability of 0.01 g and in an analytical balance (SHIMADZU®, model AUY220), with readability of 0.1 mg. The ingredients were initially manually mixed in plastic bowls previously washed and rinsed with deionized water, and then mixed again using a semi-industrial mixer (LIEME®, Caxias do Sul, Brazil), for approximately 15 minutes. The mixtures were placed in polyethylene bags, properly identified, dated, and stored at 10 °C.

2.4 Biochemical analyses
Serum hemoglobin of rats was determined according to AOAC (Association of Official Analytical Chemists, 1997) using the in vitro colorimetric diagnosis kit LABTEST (Lagoa Santa, MG, Brazil). Tail blood sample was collected, and 20 µL were mixed to 5 mL of cyanide and potassium ferricyanide solution (Drabkin’s solution). Finally, absorbance of the solution was read at 540 nm on a UV-Visible spectrophotometer (SHIMADZU UV-1601, SP, Brazil). For hematocrit determination, blood was collected directly from the tail extremity by capillarity. The hematocrit was quantified by the micromethod, following the manufacturer’s instructions (Benchtop Centrifuge, Microhematocrit, SIGMA 1-15).

2.5 Experimental design
In the repletion phase, a randomized design with a 2 × 3 factorial scheme (2 DF sources versus 3 iron levels) with 8 repetitions was used, in which the animals were divided according to the hemoglobin concentration so that the means between the groups were as close as possible.

Table 2. Chemical composition and caloric value of micronized corn bran.

<table>
<thead>
<tr>
<th>Chemical Composition</th>
<th>Mean 100g⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture (g)</td>
<td>1.59</td>
</tr>
<tr>
<td>Lipids (g)</td>
<td>5.49</td>
</tr>
<tr>
<td>Proteins (g)</td>
<td>5.50</td>
</tr>
<tr>
<td>Carbohydrates (g)</td>
<td>12.54</td>
</tr>
<tr>
<td>Total Dietary fibers (TDF) (g)</td>
<td>73.40</td>
</tr>
<tr>
<td>- Soluble Dietary fibers (SDF) (g)</td>
<td>0.67</td>
</tr>
<tr>
<td>- Insoluble Dietary fibers (IDF) (g)</td>
<td>72.73</td>
</tr>
<tr>
<td>Ash (g)</td>
<td>1.46</td>
</tr>
<tr>
<td>Calcium (mg)</td>
<td>31.27</td>
</tr>
<tr>
<td>Phosphorus (mg)</td>
<td>170</td>
</tr>
<tr>
<td>Copper (mg)</td>
<td>ND</td>
</tr>
<tr>
<td>Iron (mg)</td>
<td>2.73</td>
</tr>
<tr>
<td>Manganese (mg)</td>
<td>1.67</td>
</tr>
<tr>
<td>Potassium (mg)</td>
<td>5180</td>
</tr>
<tr>
<td>Sodium (mg)</td>
<td>1185</td>
</tr>
<tr>
<td>Zinc (mg)</td>
<td>2.49</td>
</tr>
<tr>
<td>Caloric value (kcal)</td>
<td>121.65</td>
</tr>
</tbody>
</table>

Means of triplicates, except for fibers, which were analyzed in duplicate; ND: Not detected.

The data were analyzed by analysis of variance and regression analysis. As for the qualitative variables, the means were compared by the F test, at 5% probability. As for the quantitative variables, the models were analyzed based on the significance of regression coefficients, using the F test, at 5% probability, for the determination coefficient and the phenomenon in study.

The Dunnett’s test was used to compare each experimental group with the control group. The statistical analyses were performed using the System for Statistical and Genetic Analyses (SAEG) version 9.1 and Excel.

3 Results

3.1 Micronized corn bran
Table 2 shows the data of the nutritional composition analysis of corn bran: dietary fiber (73.4%) is its main component, especially the insoluble fraction (72.7%).

The fibrous corn bran had lower IP6 + IP5 levels (1.97 µmol.g⁻¹), which are the forms that show negative effect on mineral bioavailability (Table 3).

3.2 Iron bioavailability
Table 4 shows the mean values of analyzed variables in each group and the results of analysis of variance, verifying the possible interactions between the two DF sources (cellulose or corn) and the three iron levels ingested in the diets (6, 12, or 24 mg Fe.kg⁻¹).

Weight gain during depletion, total weight gain, food intake, and consequently, food efficiency ratio (FER) were not influenced by the interaction of factors or by the isolated factors (p> 0.05). The anemic condition of rats at the end of the depletion phase was confirmed by low hemoglobin levels in all groups, with average of 5.94 g/dL. The final hemoglobin and hematocrit level results and hemoglobin uptake differed only in terms of the levels of iron ingested (p≤0.01), according to data presented in Table 4. There was no difference between the source of fiber and iron level interaction (p> 0.05), which indicates that variables with the same iron level are independent.

During the repletion period, weight gain (WGR) was influenced by the ingested iron levels (p≤0.05). The equation WGR = 38.518 + 0.504 Fe (r² = 0.1229), obtained from the linear regression analysis of the data, describes the behavior of this variable so that for each unit variation in the level of Fe, there was an increase of 0.504 g in the WGR. Finally, hemoglobin (Hb), final hematocrit (Ht), and hemoglobin gain (Hg) differed only in terms of Fe level in the diet and there was no
lipids, 2.6% proteins, 2.9% minerals, 1.2% sugars, and 54% DF in the corn pericarp fraction. The high carbohydrate and lipid levels found in the product in the present study may indicate the presence of other grain fractions (germ, tip, and endosperm) during the process of pericarp fraction separation. Ferrarini (2004) found the following mean composition in yellow corn pericarp: starch 7.3%, protein 3.7%, lipids 1%, sugars 0.3%, and ash 0.8%. However, in 100 g of corn meal, our results are similar to those reported in the food composition database of the United States Department of Agriculture (2009), in terms of TDF (79 g) and iron (2.79 mg). The contents of protein (8.36 g) and total lipids (0.92 g) in the USDA database are respectively higher and lower than those found in the present study, again indicating the possibility of the presence of other grain fractions.

Linear regression analysis was carried out and the following equations describe the behavior of these variables:

\[
FHB = 5.7861 + 0.1228 \text{Fe} \quad (r^2 = 0.4318);
\]

\[
FHT = 32.69 + 0.5926 \text{Fe} \quad (r^2 = 0.4693);
\]

\[
HBG = -0.3224 + 0.1296 \text{Fe} \quad (r^2 = 0.5511).
\]

The behavior of variables associated with the blood parameters is dose-dependent, different from what was found for weight gain in the repletion phase. This may suggest that the absorptive adaptation was efficient for avoiding that weight gain at the lowest iron level was even lower than that in the intermediary level. This efficiency was not observed for hemoglobin and hematocrit recovery. Thus, FHB, FHT, and HBG values increased due to the iron levels in the diet, as seen in Table 4.

The relative bioavailability value (RBV) was calculated aiming to evaluate the corn bran diet efficiency for promoting hemoglobin recovery comparing with that of the control diet (Figure 1). In order to that, interaction was unfolded to obtain the angular coefficient of each line. The RBV of corn bran was 104%. It means the efficiency for recovering hemoglobin levels with the corn based diet was comparable to that of the standard diet with cellulose as DF source.

### Table 4. Mean values and summary of the analysis of variance as a function of the dietary fiber source, cellulose (control), corn bran experimental, and iron levels (6, 12, or 24 mg.kg⁻¹).

<table>
<thead>
<tr>
<th>DF SOURCE AND Fe (mg.kg⁻¹) LEVELS</th>
<th>WEIGHT GAIN DEPLETION (g)</th>
<th>WEIGHT GAIN REPLETION (g)</th>
<th>WEIGHT GAIN TOTAL (g)</th>
<th>FOOD INTAKE. (g)</th>
<th>FOOD EFFICIENCY RATIO (FER)</th>
<th>FINAL HEMATOCRIT (g/dL)</th>
<th>FINAL Hb (g/dL)</th>
<th>Hb GAIN (g/dL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cellulose (6 mg.kg⁻¹)</td>
<td>98.50 ± 7.63</td>
<td>40.88 ± 13.34</td>
<td>139.38 ± 11.25</td>
<td>436.24 ± 34.39</td>
<td>0.32 ± 0.03</td>
<td>35.40 ± 1.19</td>
<td>6.54 ± 0.83</td>
<td>0.44 ± 0.83</td>
</tr>
<tr>
<td>Cellulose (12 mg.kg⁻¹)</td>
<td>93.57 ± 17.49</td>
<td>38.43 ± 9.43</td>
<td>132.00 ± 13.67</td>
<td>424.87 ± 41.02</td>
<td>0.31 ± 0.04</td>
<td>41.17 ± 3.49</td>
<td>7.36 ± 1.23</td>
<td>1.26 ± 1.28</td>
</tr>
<tr>
<td>Cellulose (24 mg.kg⁻¹)</td>
<td>91.13 ± 15.01</td>
<td>51.75 ± 8.48</td>
<td>142.88 ± 11.27</td>
<td>455.13 ± 25.50</td>
<td>0.31 ± 0.02</td>
<td>48.13 ± 2.59</td>
<td>8.80 ± 0.95</td>
<td>2.83 ± 0.91</td>
</tr>
<tr>
<td>Corn bran (6 mg.kg⁻¹)</td>
<td>91.75 ± 10.53</td>
<td>45.00 ± 9.90</td>
<td>136.75 ± 18.30</td>
<td>436.22 ± 16.88</td>
<td>0.31 ± 0.04</td>
<td>34.88 ± 5.72</td>
<td>6.14 ± 1.25</td>
<td>0.14 ± 0.93</td>
</tr>
<tr>
<td>Corn bran (12 mg.kg⁻¹)</td>
<td>102.25 ± 10.61</td>
<td>45.75 ± 7.92</td>
<td>148.00 ± 11.51</td>
<td>461.99 ± 28.86</td>
<td>0.32 ± 0.03</td>
<td>41.63 ± 7.07</td>
<td>7.64 ± 1.25</td>
<td>1.71 ± 1.71</td>
</tr>
<tr>
<td>Corn bran (24 mg.kg⁻¹)</td>
<td>97.13 ± 12.69</td>
<td>50.88 ± 12.39</td>
<td>148.00 ± 13.88</td>
<td>442.70 ± 34.22</td>
<td>0.34 ± 0.05</td>
<td>44.75 ± 4.95</td>
<td>8.51 ± 0.72</td>
<td>2.59 ± 0.77</td>
</tr>
</tbody>
</table>

Mean Squares

| Source (DF=1) | 81.86 NS | 145.54 NS | 445.72 NS | 792.56 NS | 0.00 NS | 12.2300 NS | 0.0075 NS | 0.1428 NS |
| Level (DF=2)  | 58.96 NS | 409.65 *  | 233.15 NS | 646.70 NS | 0.00 NS | 373.8000 ** | 20.8500 ** | 25.8100 ** |
| Source x Level (DF=2) | 266.88 NS | 66.29 NS | 336.96 NS | 2545.05 NS | 0.00 NS | 14.7900 NS | 0.2886 NS | 0.9853 NS |
| Bran (DF=41)  | 158.79   | 109.32    | 183.37    | 950.04    | 0.00    | 24.6600    | 1.1400    | 0.9233    |

Coefficient of variance

| 13.15 | 22.93 | 9.57 | 6.95 | 11.91 | 11.88 | 14.12 | 66.01 |

Results are expressed as mean ± SD of iron-deficient rats (n=8/group) after being fed an iron deficient diet for 21 days: the control group (cellulose) and experimental group (corn bran). The Dunnett’s test was used to compare each experimental group and the control group; * - F at 5% significance level; ** - F at 1% significance level; NS - F non-significant at 5% significance level; DF=degree of freedom.

4 Discussion

The micronized corn bran is a viable source of insoluble dietary fiber. According to Giuntini et al. (2003), a food or product with 2 to 3% of DF can be considered a good source of this nutrient. When there is a two fold increase in its content (4 to 6%), it is considered a food with high level of the nutrient; the dietary fiber content of the analyzed product was 12 times higher than that.

It is important to consider that non-conventional products and foods are potential sources of nutrients with application in human food. However, there is little information available on these products. Paes (2006) found (dry basis) 0.6% starch, 1.3% lipids, 2.6% proteins, 2.9% minerals, 1.2% sugars, and 54% DF in the corn pericarp fraction. The high carbohydrate and lipid levels found in the product in the present study may indicate the presence of other grain fractions (germ, tip, and endosperm) during the process of pericarp fraction separation. Ferrarini (2004) found the following mean composition in yellow corn pericarp: starch 7.3%, protein 3.7%, lipids 1%, sugars 0.3%, and ash 0.8%. However, in 100 g of corn meal, our results are similar to those reported in the food composition database of the United States Department of Agriculture (2009), in terms of TDF (79 g) and iron (2.79 mg). The contents of protein (8.36 g) and total lipids (0.92 g) in the USDA database are respectively higher and lower than those found in the present study, again indicating the possibility of the presence of other grain fractions.
Effects of corn bran on iron bioavailability

Besides the pericarp in the product evaluated; corn meal also has 42 mg of Ca and 1.56 mg of Zn. As reported in the literature, the negative effects DF-source foods on mineral bioavailability is associated with the phytate content and not with the fiber content itself (Lestienne et al., 2005; Schleemmer et al., 2009). According to Sandberg (2002), only IP5 and IP6 have negative effects on mineral bioavailability. The other compounds formed have low mineral binding capacity or the conjugates formed are more soluble (Lonnrdal, 2000) Therefore, the sum of these two compounds was considered. The fibrous corn bran had lower IP6 + IP5 levels (1.97 μmol.g⁻¹) when compared to those of leguminous plants such as bean (20.96 to 26.73 μmol.g⁻¹) (Ramirez-Cárdenas et al., 2008) and to the IP sum in soybean (20.15 μmol.g⁻¹) (Martino et al., 2007). As for cereals such as oat flakes (13.2 μmol.g⁻¹), milheto or Italian millet (7.91 μmol.g⁻¹), and wheat flour (6.36 μmol.g⁻¹), the values were also low (Ma et al., 2005). In the whole corn grain, Lorenz et al. (2007) found that the varieties known for having low phytate level, had 4.01 μmol.g⁻¹ on average, and those known for having high level had 5.75 μmol.g⁻² Ma et al. (2005) found a phytate concentration of 4.55 μmol.g⁻¹ in fresh corn, which is within the range 4.01-5.75. In the present study, the results of the micronized bran analysis, which is basically composed of the pericarp grain, confirm that most of the phytate is found in other parts of the corn grain, especially in the germ, , as reported by Mjoun et al. (2008).

Therefore, since the health benefits of regular consumption of dietary fibers have been widely reported in the literature (Mann & Cummings, 2009), and based on studies that show their low intake by populations of different age and gender groups in different regions, the need to find alternative dietary fiber sources that are affordable and cost-effective is confirmed; especially those with low in IP6 +IP5, such as the corn bran evaluated in the present study.

Concerning iron bioavailability, it was observed that the weight gain was influenced by the ingested iron levels (p<0.05) during the repletion period. This can be explained because iron deficiency has great influence on the weight gain of animals in the growth phase (Tinkov et al., 2012). Similar to our findings, Hamdaoui et al. (2003) investigating diets with legume, meat, black tea, and green tea found that rats which consumed iron with higher bioavailability gained more weight than the rats that consumed ferrous sulfate. Strube et al. (2002) reported lower weight gain (p<0.01) in Sprague-Dawley rats fed an iron-deficient diet. Similar behavior was found by Hernández et al. (2003) in Sprague-Dawley rats fed a wheat flour diet with low iron levels.

It was also observed that, below the recommended daily limit, weight gain does not show linearity with the level of iron. In other words, in the case of inadequate iron intake, higher intake, for example, 12 mg kg⁻¹, does not guarantee the body will get a sufficient amount of iron. This fact can probably be explained by the physiologic adaptation to low ingestions, situation in which iron absorption is increased to increase biological use of this lower quantity offered (Donker et al., 2014).

The low phytate level (Table 3) may have contributed to high iron bioavailability in the studied product since phytate interference in the bioavailability of this mineral has been widely reported in the literature (Moura & Canniatti-Brazaca, 2006; Schleemmer et al., 2009). Welch (2002) suggests that the microorganism population present in the intestine and the diet composition would play an important role in the effects of phytates on iron and zinc bioavailability; however, further investigation is necessary to clarify this possibility.

Nevertheless, this evaluation must be done according to the phytate:Fe ratio, which according to Hallberg et al. (1989), affects the bioavailability of this mineral when higher than 1. In contrast, Saha et al. (1994), studying diets containing wheat flour, reported that iron absorption in rats significantly reduced when the phytate:iron ratio was higher than 14. In the present study, the following values were found for the millimolar phytate:Fe ratios per kg of diet in the levels 6, 12 and 24 mg Fe.kg⁻¹ , 1.33, 0.63, and 0.31, respectively. For the levels 12 and 24 mg. kg⁻¹ , the phytate:Fe ratio was lower than the reference values, which can explain the similarity (p> 0.05) in the blood parameters in terms of the DF source of the diets with the same iron level. No difference was observed in the level 6 mg Fe.kg⁻¹; either, and since the ratio is higher than 1, this may indicate that the value suggested by Hallberg et al. (1989) must be higher; values higher than this will affect iron bioavailability.

Corroborating our findings, Fly & Czamecki-Maulden (1996), studying chicks evaluated the iron bioavailability in crackers and cereals rich in DF, found that moderate quantities (2-4%) of DF in corn did not affect iron bioavailability in anemic chicks. In our study, the quantity of DF was 5%. According to Fantini et al. (2008), DF does not affect mineral utilization such as iron, and whole cereals have inhibitory effect only due to the fact that they have phytate in their composition.

On the other hand, Hernández et al. (2003) reported that Sprague-Dawley rats fed a alkalai- cooked corn flour diet showed lower iron absorption and hemoglobin level than those treated with wheat flour. These authors add that this fact must have occurred because the corn flour contained almost four times more DF and phytate, and the alkaline environment may have contributed to reduce Fe absorption since in this environment, the ferric form predominates and is less absorbed than the ferrous form. According to Lin et al. (2005) and Nakajima et al. (2012), the phytate content varies considerably between different corn genotypes. Therefore, flours from different cultivars may also have different molar ratios (Queiroz et al., 2011) thus influencing mineral bioavailability, which might explains one of the differences found in their study. With regard to the negative influence of DF on mineral bioavailability (Lestienne et al., 2005), no interference was found in our study although the product being evaluated had high concentration (73.4%) of this nutrient.

The RBV value was 104%, showing efficiency for recovering hemoglobin levels. This was an interesting finding since, in general, RBV values higher than those of the control group is found when an iron absorption promoter is used, as in case of a study in Tunisia, in which the experimental group fed with a typical leguminous plant with meat had RBV = 122% (Hamdaoui et al., 2003). Therefore, this bran seems to be a good alternative as DF source.
It is known that, with 73.4% of TDF, the studied product is a major source of DF. However, the assertion that it is a major source of this nutrient, in other words, that it provides the same beneficial health effects as other DF sources (Jones, 2013), or that it is not a consequence of chemical interactions that affects the bioavailability of other nutrients, specially minerals, cannot be made yet. With regard to iron bioavailability, a negative effect on the analyzed parameters was not observed, given that hemoglobin recovery efficiency was 104% comparing with that of the control group.

It can be said that the analyzed product is a good source of insoluble DF only, but both DF types are important since they may enhance the benefits attributed to them, such as the reduction in the risks of cardiovascular diseases and type 2 diabetes (Galisteo et al., 2008).

5 Conclusion

Micronized corn bran is a viable dietary fiber source, mainly insoluble dietary fiber, contributing to the increase in the iron and zinc levels. It is a low-calorie product and has low phytate content (IP5 and IP6 fractions); it showed no negative effect on iron bioavailability in rats. Such characteristics suggest the possibility of its use as dietary fiber ingredient for the enrichment of food products. Further studies to evaluate its functional effects are needed in order to support its use as source of beneficial DF to human health.

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