Evaluation of the Tannin Content in Cereals Consumed in Chad: *Sorghum caudatum* and *Digitaria iburua*

A. Mbaiogaou, S. M. Betoloum, S. Mbaihougadobe, M. Naitormbaide, A. D. Adoum, Y. Mahmout

**ABSTRACT**

The seeds of *Sorghum caudatum* called red sorghum and *Digitaria iburua* called fonio were studied for their tannin contents. The tannin contents of these two species were evaluated by the reference method for sorghum tannins. The seed parts studied were the crushed seed set, the starch and the bran. The seeds of *Sorghum caudatum* and *Digitaria iburua* contained 1.663 mg EAG/100 g P and 0.902 mg EAG/100 g P, respectively. The tannin contents of *Digitaria iburua* and *Sorghum caudatum* brans are respectively 0.866 mg EAG/100 g P and 0.922 mg EAG/100 g P. Tannins were much more lodged in the bran part of these two species.

**Keywords:** Chad, Content, *Digitaria iburua*, *Sorghum caudatum*, Tannins.

I. INTRODUCTION

Tannins are secondary plant metabolites originally recognized because they interact strongly with collagen, converting animal skin into leather. They are high molecular weight polyphenols that precipitate proteins in solution (Haslam E., 1998).

Within the large group of natural compounds called tannins, a variety of chemical structures are possible, and the number of plant species containing these compounds in non-negligible amounts is immense (Hagerman A. E., 1998 and Haslam E., 1989). Hydrolyzable tannins consist of simple phenolic acids such as gallic acid esterified to polyols, usually glucose, and condensed tannins are polymers of flavonoid units (Stern et al., 1996).

Tannins effectively protect plants that are rich in them, against predators of all kinds (herbivores, birds, insects, molds ...), thanks to their anti-nutritional properties, astringency and poor coloring. In addition, tannins prevent
the pregermination of seeds.

These advantages are often coupled with disadvantages when the organs of the plant rich in tannins are used in food: it is in particular about the binding and the precipitation with the proteins which involves a fall of the digestibility, of bad coloration and the bad taste which they confer to the foods.

Several human staple foods, including cereals, fruits, beverages, and some seeds contain condensed and hydrolyzable tannins, leading to an estimated daily dose of one gram of tannins (Pierpoint W. S., 1990). There is a great potential for tannin-protein complexes to form in the gastrointestinal tract, given that diets and the lining of the gastrointestinal tract are filled with protein. In addition, the tannin-protein complexes prepared in vitro are stable at the pH of the gastrointestinal tract in the presence of protease enzymes (pepsin, trypsin, chymotrypsin, elastase, carboxypeptidase A, B) and bile acids (Yan Q and Bennick A., 1995 and Lu Y. and Bennick A., 1995). The increase in fecal nitrogen, which is symptomatic of the consumption of a high tannin diet, is the consequence of the formation of stable tannin-protein complexes in the gastrointestinal tract (Lu Y. and Bennick A., 1995).

Cereals such as Sorghum caudatum and Digitaria iburua are widely consumed in many parts of Chad. Africa has more than half (55%) of the world's sorghum production (Fahima et al., 2018). Currently, with breeding improvement, the number of identified sorghum varieties is more than 7,000 (Fahima et al., 2018 and Trouche et al., 1999). Sorghum caudatum processing into malt can also find application in beer industries as a replacement for barley malt if the amylase activities (especially α-amylases, β-amylases and limit dextrinases) are sufficient. It can be used also in the preparation of weaning foods (slurries) with low viscosity (Khady et al., 2010). However, the consumption of these cereals causes serious problems of digestion. This suggested that Sorghum caudatum and Digitaria iburua contain tannins.

The present purpose is to determine tannin content in the seeds of these two cereals widely consumed in Chad.

**II. MATERIALS AND METHODS**

**A. Plant Material**

The study focuses on the seeds of Sorghum caudatum, locally called red sorghum, and Digitaria iburua, called black fonio. The seeds of Sorghum caudatum and Digitaria iburua were provided by the Chadian Institute of Agronomic Research for Development of Bebedja city, Chad. The different parts studied are the crushed seed set, the bran and the starch of each of the two species. These different parts were grounded with mortar in powder.

**B. Methods**

1) **Sample Preparation and Extraction of Phenolic Compounds**

Different dry seed parts of the species were ground with a mortar. Then, 5 g of powder of each species were weighed and put in 30 mL of acetone-water system (80: 20, v/v) and kept in the refrigerator at 4°C for three days (Asami et al., 2003). Extracts were filtered and the filtrates were kept in the refrigerator for subsequent tannin determination.

2) **Method of Determination of Tannins**

We adopted the reference method for sorghum tannins to the tannins of the species studied (JOCE, 2011).

Addition of ferric ammonium citrate (III) and ammonia to part of the acetone phase and spectrophotometric measurement of the absorbance of the resulting solution at 525 nm. Determination of the tannin content using a calibration curve obtained from gallic acid as reference.

3) **Qualitative Analysis**

Tests carried out on the 5% infusion.

To 2.5 g of powdered plant material, we added 50 mL of boiling water; and we let infuse 15 minutes and filtered. We added some drops of FeCl₃ in the infused. If there is formation of green, brown or blue coloration with precipitate:
- blue black: presence of gallic tannins;
- green-brown: presence of catechin tannins (Mahmout Yaya, 1987).

Establishing the calibration curve

Establish the calibration curve as follows:
- Using a pipette, we prepared and introduced 6 test tubes and introduced with a pipette respectively: 0, 1, 2, 3, 4, and 5 mL of the aqueous solution of gallic acid such that the total volume of each tube was 10 mL.
- Finally, Pipette into six other test tubes we divided 1 mL of each of these solutions into 6 test tubes and added successively 5 mL of distilled water, 1 mL of iron (III) ammonium citrate solution and then; stirred for a few seconds using the vortex shaker.
- The calibration curve was plotted with the absorbance values on the ordinates and the corresponding concentrations on the abscissa (JOCE, 2011).

The resulting solutions were transferred to the measuring cuvettes and the absorbances were measured 10 minutes later at 525 nm against water.

4) **Quantitative Analysis**

The determination of tannins has been carried out according to the standard methods of Mahmout Yaya, 1987. Extraction with chloroform

After extraction of the phenolic compounds with chloroform, the determination is carried out. Assay

1 - Pipette 1 mL of the acetone phase into a test tube n° 1. Add successively 5 mL of distilled water and 1 mL of ammonia, then shake for a few seconds with the vortex mixer.
2 - Pipette 1 mL of the acetone phase into a test tube marked No. 2. Add successively 5mL of distilled water, 1 mL of the iron (III) ammonium citrate solution. Shake for a few seconds using the vortex mixer.
3 - Transfer the solutions n° 1 and n° 2 into the measuring cuvettes (1 cm) and measure the absorbances with a spectrophotometer at 525 nm against water, 10 min after the end of operations 1 and 2. The result is the difference in absorbances (Mahmout Yaya, 1987).

5) **Statistical Analysis**

The statistical study was performed by Excel software at the 5% probability threshold. All experiments were performed in triplicate. The results are expressed as mean ±
standard deviation. Values of p < 0.05 are considered statistically significant (Athamena et al., 2010).

III. RESULTS AND DISCUSSION

A. Results

1) Results of the Qualitative Analysis

The phytochemical screening allowed to obtain the results that are recorded in Table I.

<table>
<thead>
<tr>
<th>Material Plant</th>
<th>Results</th>
<th>Observations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ground sorghum</td>
<td>++ + +</td>
<td>Blue typically coloration</td>
</tr>
<tr>
<td>Sorghum bran</td>
<td>++</td>
<td>Light blue coloration</td>
</tr>
<tr>
<td>Ground fonio</td>
<td>++ + +</td>
<td>Blue typically coloration</td>
</tr>
<tr>
<td>Fonio bran</td>
<td>+ +</td>
<td>Light blue coloration</td>
</tr>
</tbody>
</table>

2) Results of the Qualitative Analysis

The total tannin content of the extracts was expressed in milligrams of Gallic Acid Equivalent per hundred grams of powder (mg GAE/100 g P). A calibration curve was drawn with gallic acid at different concentrations; optical density measurements for each extract were performed at 525 nm. Total tannin contents were determined according to the equation $y = 7.9851x + 0.0224$ with the correlation coefficient $R^2 = 0.9935$ (Fig. 1).

![Gallic acid calibration curve.](image)

Results of the quantitative analysis were designed in Table II below. The action of iron (III) chloride on the 5% infusion showed that the different seed parts of *Sorghum caudatum* and *Digitaria Iburua* contain tannins.

<table>
<thead>
<tr>
<th>Species</th>
<th>Plant material</th>
<th>Total tannin content (mg GAE/100g P)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Sorghum caudatum</em></td>
<td>Ground seeds</td>
<td>$1.663 \pm 0.0042a$</td>
</tr>
<tr>
<td></td>
<td>Bran</td>
<td>$0.922 \pm 0.0011b$</td>
</tr>
<tr>
<td></td>
<td>Starch</td>
<td>$0.574 \pm 0.0006c$</td>
</tr>
<tr>
<td><em>Digitaria Iburua</em></td>
<td>Ground seeds</td>
<td>$0.902 \pm 0.0011a$</td>
</tr>
<tr>
<td></td>
<td>Bran</td>
<td>$0.866 \pm 0.0006b$</td>
</tr>
<tr>
<td></td>
<td>Starch</td>
<td>$0.764 \pm 0.0008c$</td>
</tr>
</tbody>
</table>

Superintendent values with the same letters in the columns were not significantly different (p < 0.05) according to Duncan's multiple comparison test.

IV. DISCUSSION

The preliminary phytochemical tests showed that the sorghum extract has an appreciable quantity of tannins. This gives it a blue typically. These results corroborate those of Ba K., et al, 2010 who worked on the comparative study of phenolic compounds, antioxidant power of different varieties of Senegalese sorghum and amylolytic enzymes of their malt.

This traditional sorghum variety is appreciated for its grain quality, but is not very productive. Until now, the selected varieties had better yield but poor grain quality. Today, knowledge on the biochemical nature of quality and on the genetic support of these criteria has progressed considerably, opening the way to the selection of new varieties that are productive and well accepted by consumers according to G. Trouche et al., 1999.

The results of the assay revealed on the one hand that the extract of the whole ground seeds of *Sorghum caudatum* contains more tannins than that of bran (Table II, Fig. 2). On the other hand, the bran extract was richer in tannins than the starch extract. The following tannin contents were noted: 1.663, 0.922 and 0.574 mg EAG/100 g P respectively for the extracts of ground seeds, bran and starch. It can be seen that for this species, compared with starch extract and bran extract contained more tannins.

The seeds of *Digitaria Iburua* showed that extract of the ground seed contained more tannins than the extracts of bran and starch with the values 0.902, 0.866 and 0.764 mg EAG/100 g P respectively (Table II, Fig. 3). In both cases, tannins were much more accumulated in the bran parts than in the starches of both species.

The comparative study of two species showed on the one hand that the *Sorghum caudatum* extract was richer in tannins than the *Digitaria Iburua* extract for the powders of respective contents 1.663 and 0.902 mg of GGE/100 g P (Table II, Fig. 4). These values thus obtained confirm the coloration observed during the revelation test on the 5% infusion. On the other hand, for the different parts studied, the extract of *Sorghum caudatum* bran contains a lot of tannins than the extract of *Digitaria Iburua* bran with values respectively 0.922 and 0.866 mg EAG/100g P. The extract of *Sorghum caudatum* starch was less rich in tannins than that of *Digitaria Iburua* with contents respectively 0.574 and 0.764 mg EAG/100 g P.

According to the results, tannins were much more accumulated in the bran part of both species than in the starch. So, removing the bran from both species is enough to reduce the risk of binding and precipitation with proteins, which leads to a decrease in digestibility.

Tannin-rich sorghums are an important source of antioxidants which are interesting for human health. Their disadvantage is at the level of tannin-protein interactions, particularly for *Sorghum caudatum*. These interactions affect the digestibility of proteins and carbohydrates, but also inhibit enzymes like malt amylases. It should be recognized that currently many traditional foods such as porridges and alcoholic beverages are made from tannin sorghum (Awika et al., 2004).
This work allowed to highlight the presence of tannins in the seeds of *Sorghum caudatum* and *Digitaria iburua*. Thus, the seeds of *Sorghum caudatum* contain more tannins than those of *Digitaria iburua*. However, from the different parts of these two species studied, the bran extract of *Sorghum caudatum* is richer in tannins. Tannins are more accumulated in the brans of two cereals. Removal the brans from these two species is enough to reduce the risk of binding and precipitation with proteins, which leads to a decrease in digestibility. This study determined a high level of tannins in *Sorghum caudatum* and *Digitaria iburua* seed extracts so it is good to note that it is often necessary to remove the bran of these species before consumption.

REFERENCES


