

**CHARACTERIZATION AND IDENTIFICATION OF BIOACTIVE COMPOUNDS IN AGRO-FOOD WASTE FLOURS****Camila Gonçalves Rodrigues<sup>a</sup>, Viviane Dias Medeiros Silva<sup>a</sup>, Anna Cláudia de Freitas e Loyola<sup>a</sup>, Mauro Ramalho Silva<sup>a</sup>, Rodinei Augusti<sup>b</sup>, Julio Onesio Ferreira Melo<sup>c,\*</sup>, Lanamar de Almeida Carlos<sup>d</sup> e Camila Argenta Fante<sup>a</sup>**<sup>a</sup>Faculdade de Farmácia, Universidade Federal de Minas Gerais, 31270-901, Belo Horizonte – MG, Brasil<sup>b</sup>Departamento de Química, Instituto de Ciências Exatas, Universidade Federal de Minas Gerais, 31270-901, Belo Horizonte – MG, Brasil<sup>c</sup>Departamento de Ciências Exatas e Biológicas, Universidade Federal de São João del-Rei, 35702-031 São João del-Rei – MG, Brasil<sup>d</sup>Departamento de Engenharia de Alimentos, Universidade Federal de São João del-Rei, 35702-031 São João del-Rei – MG, Brasil

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One of the recurrent problems of the agroindustry sector is the incorrect disposal of waste because of the complexity of the material, moisture, among other factors. Waste is not used in tonnage, but is systematically used. Examples of residues are malt bagasse and cassava peel, produced in large quantities in the brewing industry and flour production, respectively. In this context, the objective of this study was to evaluate bioactive compounds of flours produced from agroindustry waste to potentiate their use as an ingredient rich in antioxidants. The flours produced from agroindustry waste were evaluated for phenolic compounds, total carotenoids, antioxidant activity, and the attainment of phenolic profiles and fingerprints by paper spray mass spectrometry. It was possible to identify a broad class of compounds in both flours, such as phenolic acids, flavonoids, among others. The flours can be possibly included in food commodities or packaging formulation since they are natural antioxidants, thus adding value to these products and reducing environmental issues. However, further analysis is needed to ensure bioavailability as well as food safety.

Keywords: waste; fragments; phenolics; cassava peel; malt bagasse.

**INTRODUCTION**

Waste generation in the agroindustry sector has been a recurring problem that compromises the environment. Tons of plant waste is produced annually by different industrial segments. Due to the large volume generated, transport difficulty, low added value, and high propensity for microbiological growth, these residues are destined mainly for supplementation in animal feed, used as raw material for combustion and/or disposed of in landfills. Their underutilization is due to the difficulty in treating them to be used again by the industry.<sup>1</sup> The big challenge is to provide alternative, sustainable destinations in quantity for waste.<sup>2</sup> This amount includes the by-products of the exploitation of starch sources, as well as waste from the brewing industry, which, for having peculiar characteristics, has gained space as the raw material of different commodities, at a time when the appeal for natural products by a large part of the population is increasing.<sup>3</sup>

The progressive increase in production in the brewing industry leads to many residues such as malt bagasse.<sup>4</sup> Barley malt bagasse is formed after malt germination and mashing, in the must filtration stage, generating 14 to 20 kg of this residue for every 100 liters of beer produced.<sup>5,6</sup> The destination of malt bagasse varies according to the region, and it can be directed to several applications: human food, energy production (charcoal, biogas, and direct combustion), incorporation in construction materials, paper production, adsorbents, and mainly for use as animal feed.<sup>7,8</sup>

Studies conducted by the Brazilian Institute of Geography and Statistics highlighted that in 2019 the Brazilian cassava production reached 18.9 million tons, and 60% of the harvested roots are used in flour production with a high residue generation.<sup>9,10</sup> During the various processing stages, dry and wet residues are generated, most of which are dried, discarded directly in the soil, or used as animal feed.<sup>10</sup>

Cassava peel, a material rich in fibers, minerals, and sugars, has been used as animal feed and energy production.<sup>11,12</sup> Malt bagasse contains proteins and fibers (cellulose, hemicellulose, and lignin) in its composition in addition to extractives and ash in smaller proportions.<sup>13</sup> The search for bioactive compounds comprises an appealing possibility to aggregate value to these underused residues, some studies report this possibility.<sup>14-18</sup> Bioactive compounds can promote health benefits due to their antioxidant and antibacterial activities and their capability to stimulate the immune system, among other properties.<sup>19</sup>

High-performance liquid chromatography (HPLC) has been currently used to identify bioactive compounds.<sup>20</sup> HPLC provides qualitative and quantitative analyses, but with some disadvantages, such as the need for time-consuming sample preparation and chromatographic run steps and high costs.<sup>21</sup> Recent developments in ambient ionization mass spectrometry may be an alternative to overcome these limitations, especially analysis time and reagents. These techniques have been used to obtain fingerprints of several complex matrices via ultra-fast, low-cost, and clean analyses.<sup>22,23</sup> Among ambient ionization techniques, paper spray mass spectrometry (PS-MS), developed by Wang *et al.*,<sup>24</sup> stood out for its remarkable ability to quickly and efficiently generate fingerprints from complex samples.<sup>20,25-28</sup>

The present manuscript aims to analyze, detect and identify bioactive compounds in some agroindustry residues, namely cassava peel, and malt bagasse. The prospect for their possible uses in new market segments, with the consequent reduction of their disposal in nature, is also part of the objectives of this work.

**EXPERIMENTAL**

For the production of flours, cassava peel and malt bagasse purchased at the local market in Sete Lagoas, MG, Brazil, between

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May and June 2018 were used as raw materials. For the preparation of the flours, the residues were dried in a cabin dryer with forced air circulation (Sparrow, PE 60 Model, Petrópolis, Brazil) at 70 °C for 12 hours. The dry material was crushed in a Walita Food Processor (750 W), with the grinder attachment for 2 minutes at speed 1 (9500 rpm). Cassava peel (CPF) and malt bagasse (MBF) flours were placed in amber glass jars and stored at -18 °C (in a freezer) until use, in order to avoid moisture and loss of bioactive compounds. The other reagents used were of analytical grade.

### Tannins

The detection of tannins in the flours was performed according to the vanillin/HCl method with absorbance reading in a spectrophotometer (Micronal AJX-1900 model, São Paulo) at 500 nm.<sup>29</sup> Tannin concentration in the samples was determined by building a standard curve with different catechin concentrations. The absorbance of the sample was replaced in the equation  $Y = 0.268x + 0.007$ , with  $R^2 = 0.99$ , where  $x$  is tannins concentration expressed in mg Catechin Eq/g, on dry basis and  $Y$  is the absorbance.

### Total Carotenoids

After extraction with acetone and partition with petroleum ether, the total carotenoids were quantified by spectrophotometry at 450 nm. The total carotenoid (C) content was calculated using the equation  $C = 100 [(A \times V \times 10^{-4}) / (E1\% \times M)]$ , where:  $A$  = Absorbance at 450 nm, multiplied by the dilution factor;  $V$  = Sample volume (25 mL);  $E1\%$  = Extinction coefficient of  $\beta$ -carotene in petroleum ether = 2592,  $M$  = Sample mass (g). The results were expressed in mg of carotenoids per gram of dry basis.<sup>30</sup>

### Extraction

The extraction was done according to Rufino *et al.*<sup>31</sup> to evaluate the total phenolic content, antioxidant activity, phenolic profile and to acquire the PS mass spectra. For this, 2.5 g of sample were weighed, and 10 mL of methanol/water (50:50, v/v) were added to a 50-mL Falcon tube. After 1 h at room temperature protected from the light, the tubes were centrifuged (Sigma, 2K15, Germany) at 2800 G for 22 min, and the supernatant was recovered. After, 10 mL of acetone/water (70:30, v/v) were added to the residue, with new incubation and centrifugation under the same conditions described. Both supernatants were mixed, and the volume was completed to 25 mL with distilled and deionized water.

### Total phenolic compounds and antioxidant activity

The total phenolic compounds of the flours were determined by the Folin-Ciocalteu method according to the methodology of SAHAN, CANSEV, and GULEN.<sup>32</sup> Antioxidant activity was evaluated by the methods FRAP and ABTS and DPPH according to the official method 2012.04 described by AOAC.<sup>31,33</sup>

### Phenolic profile

The determination of the profile of the main phenolic compounds present in the flours (CPF and MBF) was performed according to the chromatographic method described by Eça *et al.*,<sup>21</sup> with some modifications. The extract was filtered with a 0.22  $\mu$ m nylon syringe filter and placed in vials for automatic injection of 1  $\mu$ L in ultra-performance liquid chromatography (Waters, Acquity UPLC® Class, Milford, Massachusetts, USA) equipped with diode-array

UV detector, quaternary pump, online degasser, and automatic sampler. The data were processed using Empower® software. The chromatographic conditions were as follows: Acquity UPLC® BEH C18 column (2.1 x 100 mm; 1.7  $\mu$ m, Waters, Milford, Massachusetts, USA), a constant flow rate of 0.3 mL.min<sup>-1</sup> with two mobile phases (A = acetonitrile and B = water: formic acid, 99.75:0.25). The standards diluted in water (gallic acid, catechin, and chlorogenic acid) were run for 17 min in isocratic elution mode of 5:95 v/v (A:B). For the standards diluted in methanol (caffeic acid, ellagic acid, quercetin and rutin), the following elution gradients were applied: from 0 to 8 min = 8:92 (A:B); from 8 to 14 min = 15:85; and from 14 to 22 min = 25:75. The spectra were obtained at 253, 271, 279, 322, and 372 nm. Analytical curves were constructed for each standard within the concentration range from 13.75 to 220 ppm, obtaining  $R^2$  greater than 0.999. The results obtained were expressed in  $\mu$ g g<sup>-1</sup> of extract.

### PS-MS

The analysis of the chemical profile of the samples was performed using a LCQ Fleet mass spectrometer (ThermoScientific, São José, CA, USA) equipped with a paper spray ionization source. The flour samples were analyzed in positive and negative ionization modes. A triangle-shaped chromatographic paper (1.5 cm height and 1.0 cm width) was cut, held by copper support, and positioned 0.5 cm away from the mass spectrometer inlet. This system was connected to a high-voltage source from the mass spectrometer via a copper wire. Finally, 2.0  $\mu$ L of the flour extracts were applied to the central part of the triangle, 40.0  $\mu$ L of methanol were transferred to chromatographic paper, and the high-voltage applied. The analyses of each flour were performed in triplicate for the positive and negative ionization modes.<sup>20,34,35</sup>

The following MS conditions were applied: source voltage of +4.0 kV (positive ionization mode) and -3.0 kV (negative ionization mode); capillary tension of 40 V; transfer tube temperature of 275 °C; tube lens voltage of 120 V; and mass range of  $m/z$  100 to 1000 (positive and negative ionization modes). The ions and their fragments were identified based on data described in the literature. The collision energies used to fragment the compounds ranged from 15 to 40 mu (manufacturer unit).

### Statistical analysis

First, the normality of the results and the homogeneity of variances were evaluated. The mean results of the variables were analyzed by the Student's  $t$ -test (for normal distribution) or by the Wilcoxon-Mann-Whitney test (for no normal distribution) with a significance of 5%. All tests were performed with the assistance of the SPSS 15.0 software for Windows (SPSS Inc., USA).

## RESULTS AND DISCUSSION

The contents of condensed tannins (T), total carotenoids (C), and total phenolics (P) are shown in Table 1. Note that the content of these compounds remarkably differed between the flours. The T content was higher for CPF than MBF, while an opposite result is observed for C and P.

Tannins (T) are secondary metabolites of polyphenolic nature, responsible for the characteristics normally attributed to plants that contain high levels of these substances, such as astringency and the ability to precipitate proteins.<sup>36</sup> The higher T content in CPF, when compared to MBF, is possibly because CPF was obtained directly from washed cassava peel and subjected to the drying process. Although several antinutritional factors are attributed to tannins,

**Table 1.** Condensed tannins (T), total carotenoids (C), and total phenolics (P) in cassava peel (CPF) and malt bagasse (MBF) flours on a dry basis

Variables under study	CPF <sup>1</sup>	MBF <sup>1</sup>
T (mg CAT Eq./100g)	1066.98 ±0.06 <sup>a</sup>	48.67 ±0.21 <sup>b</sup>
C (µg carotenoid/100g)	161.65 ±44.28 <sup>b</sup>	333.85 ±15.24 <sup>a</sup>
P (mg GA Eq/100g)	230.83 ±37.17 <sup>b</sup>	549.41 ±13.53 <sup>a</sup>

<sup>1</sup>Mean value ± standard deviation. Means followed by equal letters in the same row, for the same parameters, do not differ at 5% significance level by the Student's t-test.

many studies show the beneficial effects upon ingestion in moderate amounts (10 to 40 g/kg of dry matter). They can act as a natural antioxidant, with anticarcinogenic, antihypertensive, and fungicide actions.<sup>36-38</sup>

Carotenoids, in addition to playing a fundamental role in plant metabolism, also have several effects health, such as increased immune response, inhibition of cell proliferation, and reduced risk of degenerative diseases.<sup>39,40</sup> Gonçalves *et al.*,<sup>41</sup> found 450 µg of carotenoid in 100 g of fresh white cassava, a value higher than that found in the present study arising from a cassava peel residue (Table 1). MBF (333.85 µg carotenoid/100 g) stood out compared to CPF, which may be related to the presence of other ingredients, in addition to malt, added during beer production.<sup>6</sup> These residues showed similar carotenoid contents (for MBF) when compared to those found for frozen açai pulp (0.21 to 3.84 mg/100 g) of different brands in a study conducted by dos Santos *et al.*<sup>42</sup> These data once again show the potential for using these residues as a source of bioactive compounds. The CPF and MBF flours showed significant levels of total phenolic contents (230.83 and 549.41 mg GA Eq/100 g of dry matter, respectively). These results were higher than those found in ripe 'Prata' banana peel (183.85 mg GA Eq/100 g),<sup>25</sup> thus showing that both residues are potential sources for extracting such compounds. A study conducted with malt bagasse by Da Costa<sup>43</sup> found 140 mg of GA/100 g in MBF, lower than the value reported herein (549.41 mg GA Eq/100 g of dry matter), a difference that may be related to the recipe for producing beer in each process. Socaci *et al.*,<sup>44</sup> found total phenolic contents (determined by the Folin-Ciocalteu method) ranging from 40.97 mg GA/100 g to 114.23 mg GA/100 g for malt bagasse, lower than those of the present study.

Regarding the total antioxidant activity determined by the ABTS, FRAP, and DPPH methods (Table 2), the flours showed higher values by the ABST method. There was no significant difference between the CPF and MBF flours (5% significance) only by the ABTS method. By ABTS and FRAP, the antioxidant activity was higher for MBF, while by DPPH, the highest activity was observed for CPF. These data may indicate that the compounds extracted from MBF probably contain molecules that react with ABTS and FRAP free radicals. Conversely, the CPF extract probably contains more strongly reducing molecules capable of reacting with DPPH free radicals.

Regarding antioxidant activity, the ABTS and DPPH methods were the ones that best suited the studied residues. The samples being tested are better free radical scavengers. There was no significant

difference between the residues by the ABTS method, and the values were close to those observed by Loyola *et al.*,<sup>28</sup> in flour of green banana peels dried in an oven at 70 °C. Some vegetables analyzed by Tiveron<sup>46</sup> showed lower values than those found in the residues analyzed here, such as zucchini (12.7 µmol L<sup>-1</sup> Trolox/g), beetroot (11.1 µmol L<sup>-1</sup> Trolox/g), and carrot (8.1 µmol L<sup>-1</sup> Trolox/g). Hence, the residues (CPF and MBF) showed considerable antioxidant activity by the ABTS assay, being even better than some vegetables frequently consumed.

In the method based on the free-radical scavenging principle (DPPH), CPF stood out, but its value was lower than that observed in green banana peels and higher than that found in pineapple residues.<sup>28,45</sup> (Table 2)

In the phenolic profile (Table 3), gallic acid was found in high amounts in both flours, followed by catechin, rutin, chlorogenic acid, and caffeic acid. Ellagic acid and quercetin were not detected in any of the two flours studied, and in MBF only gallic and caffeic acids were detected. The extraction yield of phenolic compounds correlate with various factors, such as the extraction method and solvent used.<sup>47</sup> This fact explains why some compounds were not determined by chromatography but were detected by PS-MS.

#### Full scan PS-MS (fingerprints)

Figure 1 exemplifies PS-MS spectra of the flours (CPF and MBF) in their positive and negative modes, providing information on the samples' chemical composition. The results reveal the presence of organic acids, phenolic compounds, flavonoids, quinones, triterpenoids, amino acids, sugars, and other ionizable compounds.

#### Negative mode (-)

Table 4 displays the possible compounds detected in the negative ionization mode for the CPF and MBF flours: organic acids, phenolic compounds, and flavonoids.

#### Organic acids

Cassava peel flour showed more ions classified as organic acids compared to malt bagasse flour. This difference probably occurs because the malt residue goes through more industrial processes than cassava peel, which only goes through the cleaning, sanitizing, and peeling processes. In MBF, octadecanedioic acid (*m/z* 313) was possibly detected based on the ion of *m/z* 295 arising upon its mass-selection and fragmentation. For CPF, eight organic acids were identified, including some fatty acids and citric acid. The ion of *m/z* 353, with a fragment of *m/z* 179, was identified as chlorogenic acid. Citric (*m/z* 191) and malic (*m/z* 133) acids were also observed in the PS(-)-MS, and they have also been found in studies with green and ripe banana peel residue flours.<sup>25,28</sup>

#### Phenolic Compounds

The ions of *m/z* 170, with a fragment of *m/z* 125, and *m/z* 179, with a fragment of *m/z* 135,<sup>47</sup> correspond to gallic acid and caffeic acid, respectively, were also detected in both flours. In CPF, the following phenolic acids were identified: ellagic acid (*m/z* 302 and

**Table 2.** Antioxidant activity by ABTS, FRAP, and DPPH methods in CPF and MBF on a dry basis (DB)

Method	CPF <sup>1</sup>	MBF <sup>1</sup>	Green banana peel (Oven 70 °C) <sup>28</sup>	Pineapple Residue <sup>45</sup>
ABTS (µM Trolox/g)	26.31 ± 2.68 <sup>a</sup>	35.73 ± 13.3 <sup>a</sup>	20.84	Nd
FRAP (µM ferrous sulfate/g)	0.02 ±0.01 <sup>b</sup>	0.32 ± 0.12 <sup>a</sup>	Nd	72.63
DPPH (µM Trolox/g)	24.13 ± 0.73 <sup>a</sup>	4.32 ± 0.64 <sup>b</sup>	66.61	5.63

<sup>1</sup>Average value ± standard deviation. Means followed by equal letters in the same row, for the same parameters, do not differ at 5% significance level by the Student's t-test.

**Table 3.** Phenolic profile (mg/100 g on dry basis) in cassava peel (CPF) and malt bagasse (MBF) residue flours

Phenolics	CPF <sup>1</sup>	MBF <sup>1</sup>
Gallic Acid	270.62 ± 1.13 <sup>a</sup>	134.73 ± 1.07 <sup>b</sup>
Catechin	14.87 ± 1.04	ND
Chlorogenic Acid	0.79 ± 0.42	ND
Caffeic Acid	0.19 ± 0.26 <sup>a</sup>	0.03 ± 0.02 <sup>a</sup>
Rutin	0.96 ± 0.82	ND
Ellagic Acid	ND	ND
Quercetin	ND	ND

<sup>1</sup>Average value ± standard deviation. Means followed by equal letters in the same row, for the same parameters, do not differ at 5% significance level by the Student's t-test. ND: Not detected.

fragment of  $m/z$  284) and ferulic acid ( $m/z$  193 and fragment of  $m/z$  134). Note that the chromatographic method used could not detect ellagic acid for this study (Table 3).

### Flavonoids

The flavonoid group contains the highest number of substances identified in the negative ionization mode. Flavonoids are secondary metabolites of plants, and their intake is linked to preventive action against chronic non-communicable diseases, with antioxidant, anti-inflammatory and anticarcinogenic potential and cardiovascular protection capacity.<sup>48</sup> In MBF, the ion of  $m/z$  431 was identified as vitexin based on its main fragment ( $m/z$  311). For CPF, additional flavonoids were identified: catechin ( $m/z$  289 and fragment of  $m/z$  245), quercetin ( $m/z$  301 and fragment of  $m/z$  151), and rutin ( $m/z$  609 and fragment of  $m/z$  301). Flours obtained from green banana peel residue and loquat leaf extract also contained rutin and catechin.<sup>28,35</sup> Rutin and quercetin have pharmacological properties such as anti-inflammatory action and antioxidant power, acting on free radicals responsible for degenerative diseases and cell aging and death.<sup>49</sup>

### Positive mode (+)

Table 5 presents the possible compounds detected in the positive ionization mode for the MBF and CPF flours.

### Amino acids

CPF showed the presence of L-arginine, identified in its protonated form of  $m/z$  175, with a fragment of  $m/z$  70.<sup>20</sup>

### Flavonoids

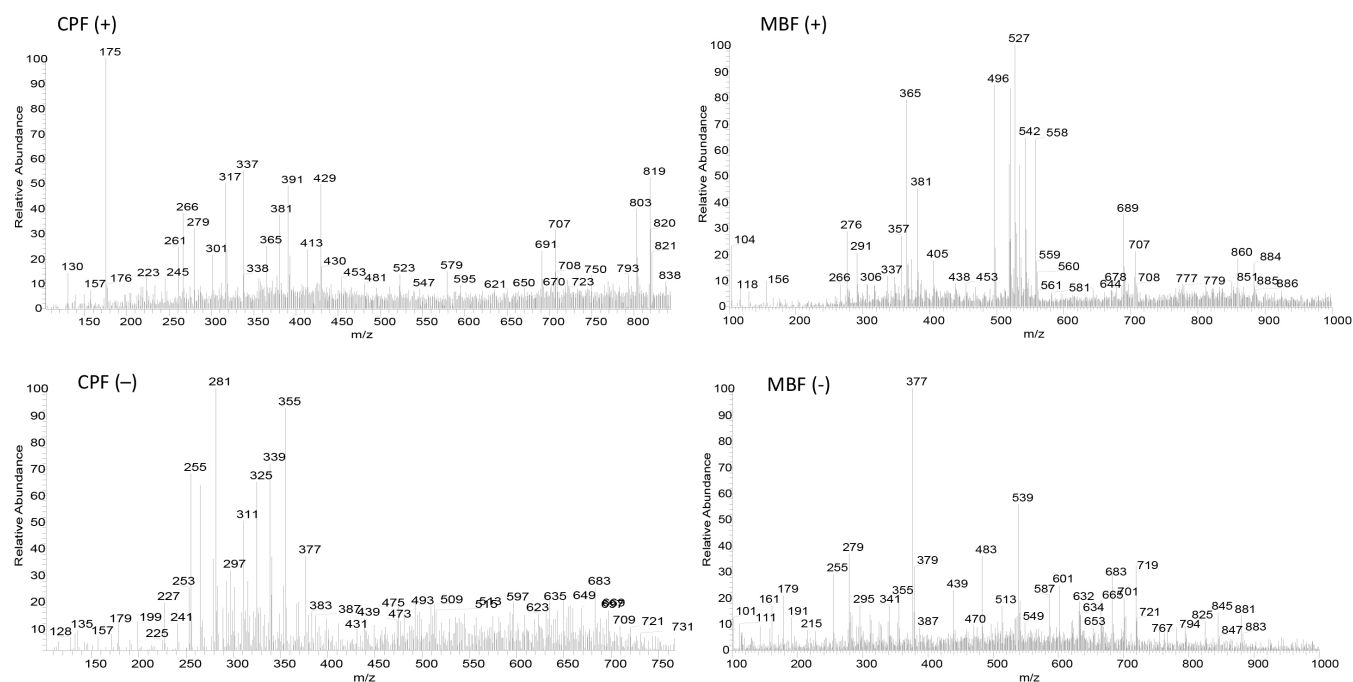
Only CPF showed molecules that could be identified as flavonoids in the positive mode. The ion of  $m/z$  595 (fragment of  $m/z$  287) was identified as tiliroside (kaempferol-3-*O*-(6-*p*-coumaroyl)-glycoside), a glycosidic flavonoid with anti-inflammatory and antioxidant properties.<sup>50</sup>

### Sugars

From the profile of ions displayed in Table 5, the ion of  $m/z$  381, observed exclusively in the PS(+)-MS of CPF, was identified as the adduct of potassium of sucrose.

## CONCLUSIONS

Even though they are agro-industrial residues, considered as products of low added value, the two flours evaluated herein (CPF and MBF) showed remarkable antioxidant activities by the ABTS method. In CPF, the obtained values of phenolic acids (gallic acid, chlorogenic acid, caffeic acid, rutin, and catechin) were higher than those found in MBF. The same trend was revealed by PS-MS data, which indicated organic acids, sugar, and phenolic acids, among other compounds, in the composition of such flours. The PS-MS method proved to be a fast and straightforward technique to obtain the fingerprint of such residues, enabling the identification of several compounds confirmed by high-performance liquid chromatography analyses described in the literature. These residues have the potential for application in the food industry, given the presence of bioactive compounds. However, further studies are required regarding the safety and bioavailability of these active compounds found.



**Figure 1.** PS-MS in the positive (+) and negative (-) ion modes

**Table 4.** Identification of ions detected in the PS(-)-MS of cassava peel (CPF) and malt bagasse (MBF) flours

Attempted identification	<i>m/z</i>	MS/MS	Reference	Flour	
				Cassava	Malt
<b>Organic acids</b>					
Malic acid	133	115	Abu-reidah <i>et al.</i> <sup>51</sup>	+	-
Citric acid	191	111	Cheikhrouhou and Khemakhem, <sup>52</sup> Silva <i>et al.</i> <sup>20</sup>	+	-
Traumatic acid	227	83	Wang <i>et al.</i> <sup>53</sup>	+	-
Palmitic acid	255	237	Zhang <i>et al.</i> <sup>54</sup>	+	-
Oleic Acid	281	237	Wang <i>et al.</i> <sup>53</sup>	+	-
Ricinoleic acid	297	183	Zhang <i>et al.</i> <sup>54</sup>	+	-
Octadecanedioic acid	313	295	Zhang <i>et al.</i> <sup>54</sup>	-	+
<b>Phenolic Compounds</b>					
<u>Benzoic acids</u>					
Gallic acid	170	125	Ee <i>et al.</i> <sup>55</sup>	+	+
Ellagic acid	302	284	Ee <i>et al.</i> <sup>55</sup>	+	-
<u>Cinnamic acids</u>					
Caffeic acid	179	135	Kang <i>et al.</i> <sup>56</sup>	+	+
Ferulic Acid	193	134	Zhang <i>et al.</i> <sup>49</sup>	+	-
1-O-Dihydrocaffeoylglycerol	255	135, 181	Kang <i>et al.</i> <sup>56</sup>	+	-
Caffeic acid hexose	341	179	Kang <i>et al.</i> <sup>56</sup>	-	+
Ferulic acid glucoside	355	193	Svarc-gaj <sup>57</sup>	+	-
Hydroxycinnamic acid derivative	379	185	Valérie <i>et al.</i> <sup>58</sup>	-	+
Coumaric acid derivative	487	145	Laura M. <i>et al.</i> <sup>59</sup>	+	-
Chlorogenic acid	353	179	Ammerer, Arle and Chieber <sup>60</sup>	+	-
<b>Quinones</b>					
Spinochrome A	265	235	Abu-reidah <i>et al.</i> <sup>51</sup>	+	-
<b>Flavonoids</b>					
Catechin	289	245	Valls <i>et al.</i> <sup>61</sup>	+	-
Quercetin	301	151, 178	Wang <i>et al.</i> <sup>53</sup>	+	-
Epigallocatechin	305	261, 287	Valls <i>et al.</i> <sup>61</sup>	+	-
Vitexin	431	311	Simirgiotis <i>et al.</i> <sup>62</sup>	-	+
Apigenin-7-O-glucoside	431	269	Svarc-gaj <sup>57</sup>	-	+
Apigenin-6-C-glucoside	431	269, 311, 341	Kang <i>et al.</i> <sup>56</sup>	-	+
9,10-Dihydro-10-(4-hydroxyphenyl)pyrano[2,3-h]epicatechin-8-on	435	189, 341	Zhang <i>et al.</i> <sup>54</sup>	-	+
Dihydrokaempferol hexoside	449	243, 405	Kang <i>et al.</i> <sup>56</sup>	+	-
Procyanidin B	577	451, 425, 407, 289	Svarc-gaj <sup>57</sup>	+	-
Rutin	609	301	Valérie <i>et al.</i> <sup>58</sup>	+	-
Apigenin-5-O-β-D-glucopyranosyl, 8-C-(6'' acetyl)-β-D-glucopyranoside	635	473	Simirgiotis <i>et al.</i> <sup>62</sup>	+	-
<b>Triterpenoids</b>					
Asiatic acid	487	469	Zhang <i>et al.</i> <sup>54</sup>	+	-
<b>Other</b>					
9-Hode	295	171, 277	Wang <i>et al.</i> <sup>53</sup>	-	+

Legend: + found, - not found.

**Table 5.** Identification of ions detected in the PS(+)-MS of cassava peel (CPF) and malt bagasse (MBF) flours

Attempted identification	<i>m/z</i>	MS/MS	Reference	Flour	
				Cassava	Malt
<b>Amino acids</b>					
L-Arginine	175	70	Silva <i>et al.</i> <sup>20</sup>	+	-
Phenolic Compounds (benzoic acid)					
Galloyl pyrogallol	279	153	Abu-reidah <i>et al.</i> <sup>51</sup>	+	-
<b>Flavonoids</b>					
Diosmetin	301	258; 286	Zhang <i>et al.</i> <sup>54</sup>	+	-
Myricetin 3-O-galactoside	481	319	Lin and Harnly <sup>63</sup>	+	-
Chrysoeriol-hexose	579	301	Abu-reidah <i>et al.</i> <sup>51</sup>	+	-
malvidin 3-(malonoyl)glucoside pelargonidin 3-rutinoside	579	433	Rio <sup>64</sup>	+	-
cyanidin 3-rutinoside	595	287	Rio <sup>64</sup>	+	-
Kaempferol rutinoside	595	287	Abu-reidah <i>et al.</i> <sup>51</sup>	+	-
Pelargonidin-3-O-(6caffeoyl-glucoiside)	595	433, 313	Wang <i>et al.</i> <sup>53</sup>	+	-
Tiliroside	595	287	Zhang <i>et al.</i> <sup>54</sup>	+	-
<b>Sugars</b>					
Sucrose	381	201, 219	Silva <i>et al.</i> <sup>20</sup>	+	-
<b>Other</b>					
Dihydroisovaltrate	424	281, 365	Abu-reidah <i>et al.</i> <sup>51</sup>	-	+
Verbonol	453	435	Abu-reidah <i>et al.</i> <sup>51</sup>	+	-
Sespendole	520	184	Abu-reidah <i>et al.</i> <sup>51</sup>	-	+

Legend: + found, - not found.

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