



## Antioxidant properties of sterilized yacon (*Smallanthus sonchifolius*) tuber flour



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### ABSTRACT

The objective of this research work was to investigate the antioxidant properties of sterilized yacon tuber flour. The results revealed for the first time the high antioxidant activity of sterilized yacon flour. The best extract obtained by boiling 8.9% (w/v) of yacon flour in deionised water for 10 min exhibited a total antioxidant capacity of  $222 \pm 2$  mg (ascorbic acid equivalent)/100 g DW and a total polyphenol content of  $275 \pm 3$  mg (gallic acid equivalent)/100 g DW associated to the presence of four main phenolic compounds: chlorogenic acid, caffeic acid, coumaric acid and protocatechuic acid, as well as the amino acid tryptophan. The most abundant was chlorogenic acid, followed by caffeic acid. Biological assays revealed that the extract had indeed antioxidant protection, and no pro-oxidant activity. In conclusion, sterilized yacon tuber flour has the potential to be used in the food industry as a food ingredient to produce functional food products.

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### 1. Introduction

Yacon [*Smallanthus sonchifolius* (Poepp. and Endl.) H. Robinson] has long been an important economic species grown for its juicy tuberous root in South America, since ancient times, on the eastern slopes of the Andes from Venezuela to north western Argentina. It was a traditional crop of the original Peruvian populations where it is still used in folk medicine. Its tubers were used for centuries as a traditional folk treatment to control hyperglycemia, kidney problems and for skin rejuvenation. Yacon is already on the European market as a prospective functional food and dietary supplement, mainly for use in certain risk groups of the general population e.g. seniors, diabetics and postmenopausal women (Valentová & Ulrichová, 2003).

Oxidative stress is widely recognized as a central feature of many chronic diseases, such as cardiovascular diseases (Jones, 2006), neurodegenerative diseases (Lin & Beal, 2006) and diabetes (Jones, 2006). As natural dietary antioxidants, phenolic compounds from plants may protect cell membranes against damage mediated by oxygen radicals (Yan et al., 1999). Research on the effects of

dietary polyphenols on human health has developed considerably. It strongly supports a role for polyphenols in the prevention of degenerative diseases, particularly cardiovascular diseases and cancers. The antioxidant properties of polyphenols have been widely studied, but it has become clear that the mechanisms of action of polyphenols go beyond the modulation of oxidative stress (Scalbert, Johnson, & Saltmarsh, 2005). Halliwell, Aeschbach, Loliger, and Aruoma (1995) defined antioxidants as “any substance that when present at low concentrations compared to those of an oxidizable substrate significantly delays or prevents oxidation of that substrate”.

Several research works (Liu et al., 2011; Zheng & Wang, 2001) established a correlation between phenolic compound content and antioxidant activity, but, in many cases, there was also antioxidant activity in the herbs that may be attributable to other unidentified substances or to synergistic interactions. Consequently, the antioxidant activity of yacon leaves extracts could therefore be predicted, to a certain extent, on the basis of phenolic compound content and profile (Valentová, Cvak, Muck, Ulrichová, & Simanek, 2003; Valentová, Šeršeň, & Ulrichová, 2005).

Five caffeic acid derivatives as main water-soluble phenolics have been isolated from yacon roots. These were identified as chlorogenic (3-caffeoylquinic), 3,5-dicaffeoylquinic acids and three caffeic and altraric acids esters (2,4 or 3,5-dicaffeoylaltraric,

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2,5-dicaffeoylaltaric and 2,3,5 or 2,4,5-tricaffeoylaltaric acids) (Takenaka et al., 2003). Takenaka and Ono (2003) also isolated derivatives of octulosonic acid from yacon tubers. Quercetin and two other flavonoids have also been identified (Simonovska, Vovk, Andrenšek, Valentová, & Ulrichová, 2003). Simonovska et al. (2003) detected chlorogenic, ferulic, and caffeic acids, quercetin, and unidentified flavonoids in yacon roots (Neves & Aparecida da Silva, 2007). A non-identified derivative of chlorogenic acid (Mr = 562) and an unknown flavonoid have also been reported (Valentová & Ulrichová, 2003).

To the best of our knowledge, no studies have been conducted to study the antioxidant activity of the yacon tubers, much less of sterilized yacon tuber flour. The studies dealing with the antioxidant potential of yacon tubers were focused on the identification and purification of the antioxidant compounds. Considering this, the objective of this research work was to study the antioxidant properties of sterilized yacon tuber flour associated to its phenolic constitution that will provide meaningful information to support its application as a functional ingredient.

## 2. Materials and methods

### 2.1. Sterilized yacon tuber flour samples

Yacon tuberous roots were obtained directly from the local producers at Campinas (SP) city, Brazil. Afterward, they were carefully washed under running water to soil removal and decontamination. Then the tubers were packed in cotton fiber bags and autoclaved at 121 °C for 20 min for enzymes inactivation and destruction of spores. Subsequently, sterilized samples were cooled down at room temperature, peeled, cross-cut and homogenised in an industrial liquefier. Next, they were packed in inox trays, frozen and lyophilized. The lyophilized material was then mashed in a mortar, thus obtaining sterilized yacon tuber flour samples for this work. Its composition expressed on a dry-weight basis (g/100 g), includes approximately, protein (2.60), lipids (0.61), carbohydrates (89.01) and ashes (3.73).

### 2.2. Extraction procedure and selection of best conditions

In order to select for the best extraction technique to obtain an extract characterized by the highest antioxidant activity, several different extraction conditions were tested – viz. temperature, stirring speed, time, solvents and  $\text{weight}_{\text{yacon}}/\text{volume}_{\text{solvent}}$  ratios.

Firstly, stirring speed, time and solvents were evaluated, using 0.4% (w/v) yacon on a dry-weight (DW) basis (Fig. 1-I), by measuring total antioxidant capacity. The solvents tested were deionised water, methanol (Methanol 205, 99.9% – gradient quality, from Romil, Great-Britain) and ethyl acetate (99.5% p.a., from Merck), at 100%. These were tested (in triplicate) by: (1) magnetic stirring at 500 rpm for 30 min and (2) 300 rpm for 20 h. Subsequently, the best solvent (deionised water), was also tested (in triplicate) (Fig. 1-II) with (3) magnetic stirring at 300 rpm for 5 min followed by 30 min sonication (model Sonorex Super RK106, from Bandelin, Germany), (4) 10 min infusion and 10 and 20 min boiling (decoction), using a condenser column.

Once the best extraction conditions were selected (those that led to higher antioxidant activity and phenolic compounds) several  $\text{W}_{\text{yacon}}/\text{V}_{\text{solvent}}$  ratios were further tested (Fig. 1-III), by determining total antioxidant capacity and total polyphenol content. The ratios, on a DW basis, tested (in triplicate) were: 3.5%, 8.9%, 13.3%, 17.7% and 26.6% (w/v) and extraction was performed for 10 min in boiling water. In order to compare the extraction efficiency of the different extraction conditions, results of antioxidant activities

were normalized as mg in ascorbic acid (equivalent)/100 g yacon tuber flour (DW).

All extracts were filtered through n° 1 filter paper (V. Reis, Portugal) and subsequent tests were carried out with the extract that demonstrated the highest antioxidant activity, i.e. extract obtained from boiling 8.9% (w/v) yacon (on a DW basis) in deionised water for 10 min.

### 2.3. Antioxidant activity

#### 2.3.1. Total antioxidant capacity

An improved 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS)-based method implemented by Gião et al. (2007) was used to determine total antioxidant capacity. According to this technique, direct production of the (blue/green) 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt radical cation (ABTS<sup>•+</sup>) chromophore was achieved via reaction between ABTS and potassium persulfate; this method is able to quantify both water and lipid-soluble antioxidants, as pure compounds or in crude extracts containing them.

The total antioxidant capacity was expressed as percentage of inhibition (PI), according to the equation  $PI = ((\text{Abs}_{\text{ABTS}^{•+}} - \text{Abs}_{\text{sample}}) / \text{Abs}_{\text{ABTS}^{•+}}) \times 100$ , where  $\text{Abs}_{\text{ABTS}^{•+}}$  denotes the initial absorbance of diluted ABTS<sup>•+</sup>, and  $\text{Abs}_{\text{sample}}$  denotes the absorbance of the sample by 6 min of reaction. Using the calibration curves, previously prepared with ascorbic acid as standard in the range 20–400 mg/L, the final result was thus expressed as equivalent concentration to ascorbic acid mg/100 g. The average of three replicates was used as a datum point.

#### 2.3.2. Total phenolic content

The total concentration of phenolic compounds was determined as described by Singleton and Rossi (1965). The chromophore development reaction is based on oxidation of polyphenols via Folin–Ciocalteu reagent (Merck), which is a mixture of phosphomolybdic and phosphotungstic acids, in a basic medium; the blue complex thus formed is assayed for absorbance at 750 nm, which is directly proportional to the total amount of polyphenols in the medium.

The total phenol content was reported as gallic acid equivalent (C; in g/L), using the expression  $C = (\text{Abs}_{\text{sample}} - 0.0201) / 2.1456$ , where  $\text{Abs}_{\text{sample}}$  denotes absorbance of the sample at 1 h of reaction. The linear regression coefficient of the above fit was 0.9991. The average of two replicates was used as a datum point.

#### 2.3.3. Phenolic composition – LC–MS/MS (liquid chromatography–mass spectrometry/mass spectrometry)

The composition in phenolic compounds of the extract with highest antioxidant activity was determined by LC–MS/MS according to the method of Politi, Rodrigues, Gião, Pintado, and Castro (2008).

Identification was based on LC and MS–MS library containing 32 phenolic compounds established by ESI–LC/MS under negative ion mode and on the literature results (Sanchez-Rabaneda et al., 2003; Sun, Liang, Bin, Li, & Duan, 2007).

In addition to our aqueous extract, a methanolic extract 100% (v/v), with the same  $\text{weight}_{\text{yacon}}/\text{volume}_{\text{solvent}}$  ratio (8.9% (w/v), DW) and magnetically stirred at 300 rpm for 1 h, was also analyzed to assess the possible presence of antioxidants that were not extracted by the water as solvent. Both extracts were prepared and injected in duplicate.

#### 2.3.4. Antioxidant activity determined via biological assays

The antioxidant activity was also tested by two distinct biological methods: (1) DNA assay by agarose gel electrophoresis, aiming

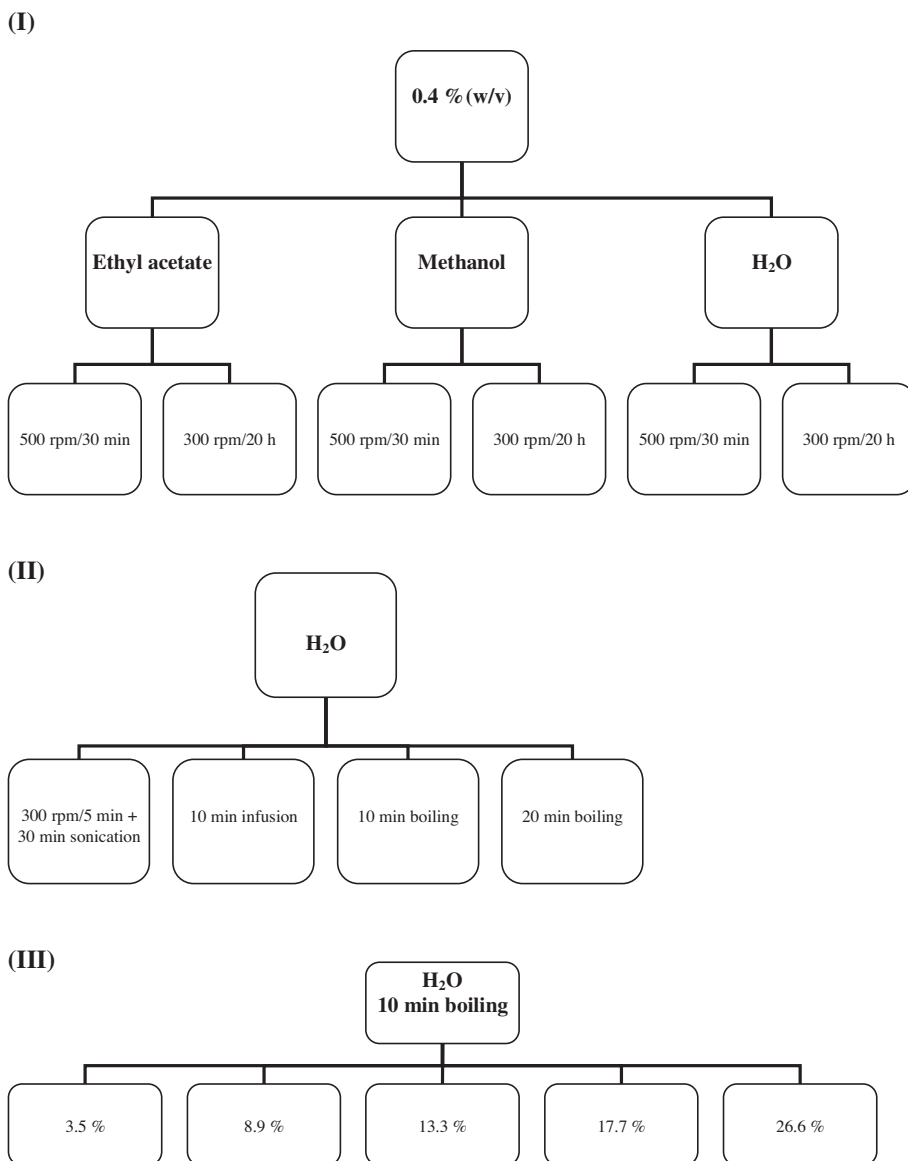


Fig. 1. Extraction procedures organigram (stirring speed, time and solvents – I and II;  $w_{\text{yacon}}/V_{\text{solvent}}$  ratios – III).

to determine direct effect of protection upon DNA; (2) bacteriophages assay, where the protective effect upon phagic DNA was determined.

**2.3.4.1. DNA assay by agarose gel electrophoresis.** The direct effect on DNA was assessed using the method described by [Gião et al. \(2008\)](#), with slight modifications. This method allows assessing both the anti- and pro-oxidant capacities of a given extract. A 400- $\mu\text{L}$  aliquot of the sample was initially prepared and processed according to the method described by [Gião et al. \(2008\)](#), with a DNA (calf thymus DNA, from Sigma–Aldrich) solution of 0.25 mg/mL.

From the resulting mixtures, 10- $\mu\text{L}$  aliquots were mixed with 1  $\mu\text{L}$  of loading buffer for DNA and loaded into the sample wells of 1% (w/v) agarose (agarose type I-A, from Sigma–Aldrich) gel, prepared with *tris*–EDTA buffer (TAE) 1% (v/v) (Sigma–Aldrich) and 10  $\mu\text{L}$  of ethidium bromide 10 mg/mL (Sigma–Aldrich). Electrophoresis was run using a power supply (model 1000/500 from Bio-Rad) at 100 V and 500 mA. DNA bands were finally digitalised using Gel Doc 2000 (Bio-Rad), with Quantity One

4.5.2, 1-D Analysis Software (Bio-Rad). Analysis was performed in duplicate.

**2.3.4.2. Bacteriophages assay.** The effect of yacon extract on phagic DNA was determined, according to the methodology described by [Gião et al. \(2009\)](#). Determination of oxidant effect upon phage, antioxidant effect upon phage with oxidant and effect of sample upon phage was performed in order to establish the potential antioxidant and pro-oxidant effect of yacon extracts.

#### 2.4. Statistical analyses

Statistical analyses were performed using SPSS 17.0 software (SPSS, Chicago, IL, USA).

In the antioxidants extraction procedures, the homoscedasticity assumption was met, hence analysis of variance (ANOVA) was applied to assess differences in the main parameters tested and their interactions, and the Tukey test was used for pair-wise comparisons, both with a 95% confidence interval. Correlation between

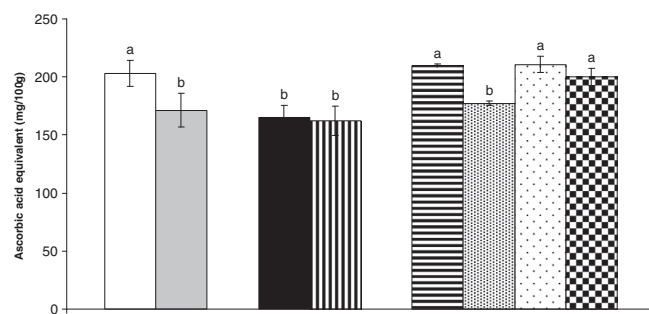
the parameters was determined by Spearman's rho, with a 99% confidence interval.

### 3. Results and discussion

#### 3.1. Extraction procedure and selection of best conditions

In order to compare the extraction efficiency of the different extraction conditions tested (stirring speed, time and solvents) on 0.4% (w/v) yacon, on a DW basis, results of the antioxidant activity were normalized as mg of ascorbic acid equivalent per 100 g yacon tuber flour (DW) and are given in Fig. 2. As can be seen, total antioxidant capacity of the different extracts obtained showed significant differences ( $p < 0.05$ ).

Ethyl acetate was the least efficient solvent extracting antioxidant compounds, since no antioxidant activity was recorded for this extract, independently of the stirring speed and time period employed. Methanol extraction led to extracts with lower antioxidant activity than those obtained with water ( $p < 0.05$ ) for the same conditions of stirring speed and time period (Fig. 2). These results, referring to extraction solvent effect upon antioxidant activity of extracts, are in agreement with the results obtained by Bravo, Monente, Juárez, Paz de Peña, and Cid (2011) for spent coffee, where among water, ethanol and methanol and their mixtures, extracts obtained with water showed the highest antioxidant capacity. However, in other studies by Lapornik, Prošek, and Wondra (2005), where a comparison of extracts prepared from plant by-products (black currant, red currant and grape marc) using different solvents (viz. water, 70% ethanol and 70% methanol) was performed, extraction with water presented the worst results regarding antioxidant capacity. In our case, water was indeed the best extraction solvent, yet its extraction capacity is affected by the combined effect of stirring speed and time period; a lower stirring speed (300 rpm) combined with a longer extraction time period (20 h) originated an extract with lower antioxidant capacity ( $p < 0.05$ ); however, when sonication was applied after a previous stirring at 300 rpm for 5 min the extraction response was significantly improved ( $p < 0.05$ ). This effect of extraction time is in agreement with the results of Lapornik et al. (2005) who found that the antioxidant activity of their extracts diminished with an increasing extraction time period. Indeed, although a longer extraction time may provide sufficient time for solute exposure to solvent and consequently enhance antioxidant compounds extraction too long a period may actually lead to antioxidant compound oxidation. For example, according to Naczk and Shahidi (2006) hypothesis, prolonged extraction times increase the chance of antioxidant compounds' degradation thus decreasing the antioxidant activity of the extract. Furthermore, from an economical perspective long extraction procedures are not desirable for industrial processes. Water extraction efficiency was also compared under different conditions and boiling showed better performance than infusion, yet a longer boiling time (20 min vs 10 min) did not improve the extraction efficiency of antioxidant compounds ( $p > 0.05$ ) neither did it reduce antioxidant capacity (Fig. 2). High temperatures may promote more efficient antioxidant compound extraction, not only because of improved solubility in the solvent but also due to alterations in the diffusion rate. Heat will disrupt more easily the interactions between phenolics and cellular constituents of plant tissues (Al-Farsi & Lee, 2008). These findings regarding the extraction method, are in agreement with those by Gião et al. (2007) concerning antioxidant extracts from medicinal plants leaves who also obtained better results when boiling, as opposed to infusion. Valentová et al. (2005), working with yacon leaves, also obtained better results, using DPPH and superoxide radicals scavenging methods to monitor antioxidant

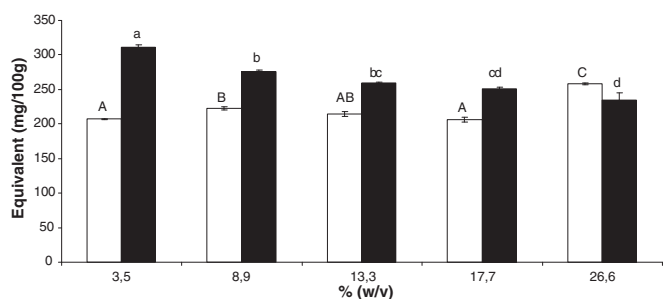


**Fig. 2.** Total antioxidant capacity of the extracts obtained with several extraction conditions of 0.4% (w/v) yacon, on a dry weight basis (extracts marked with the same letter are not significantly different ( $p > 0.05$ ;  $n = 3$ )). H<sub>2</sub>O 500 rpm/30 min □; MetOH 500 rpm/30 min ▒; H<sub>2</sub>O 300 rpm/20 h ■; MetOH 300 rpm/20 h ▨; H<sub>2</sub>O 300 rpm/5 min + 30 min sonication ▤; H<sub>2</sub>O 10 min infusion ▥; H<sub>2</sub>O 10 min boiling ▦; H<sub>2</sub>O 20 min boiling ▧. Ethyl acetate 500 rpm/30 min and ethyl acetate 300 rpm/20 h did not show any antioxidant capacity.

capacity, when boiling was used as the extraction method. However, hydroxyl radical scavenging activity was higher for infusion than for boiling. Similar results were also reported by Simonovska et al. (2003) study, where yacon leaves infusion presented higher total phenolic content than extracts obtained by boiling process. These contrasting results can be related to the different matrix characteristics and the different polyphenol distribution throughout plant parts (roots, leaves, tubers).

Total antioxidant capacity and total polyphenol content of all extracts obtained with different weight<sub>yacon</sub>/volume<sub>solvent</sub> proportions were compared (Fig. 3), since, as previously mentioned, the solvent-to-sample ratio has an effect on extraction efficiency (Naczk & Shahidi, 2006). The results reporting total antioxidant capacity and total phenolic content showed a strong linear positive correlation with each other (Spearman's rho of 0.974), which is in agreement with other research works (Campos et al., 2012; Gião et al., 2007) and implies that most antioxidant activity of the product is directly related with its phenolic content. Statistical analysis of the results (see Fig. 3) showed that there were significant differences ( $p < 0.05$ ) between the different proportions tested. Analysis of results showed that the lowest proportion that led to the highest antioxidant activity and total polyphenol content was 8.9% (w/v).

Based on the above considerations the conditions that provided the best efficiency to produce a yacon extract with the highest antioxidant activity and phenolic compounds content was that obtained by boiling 8.9% (w/v) of yacon, on a dry weight basis, in deionised water for 10 min. This extract exhibited (Fig. 3) a total antioxidant capacity of  $222 \pm 2$  mg (ascorbic acid equivalent)/100 g DW and a total polyphenol content of  $275 \pm 3$  mg (gallic acid equivalent)/100 g DW, and so this was the extract selected to carry out all the subsequent tests. Although the 3.5% (w/v) extract presented higher phenolic content, the 8.9% (w/v) was selected in order to obtain a commitment between total antioxidant capacity and total polyphenol content. The studies available dealing with the antioxidant potential of yacon tubers only established the identification and purification of the antioxidant compounds (Simonovska et al., 2003; Takenaka & Ono, 2003; Takenaka et al., 2003; Yan et al., 1999), except for the study performed by Campos et al. (2012), who determined an antioxidant capacity of 35 accessions of yacon fresh roots in the range of 0.23–13.6 mmol (trolox equivalent)/100 g DW. Regarding total polyphenol content of yacon tubers, Simonovska et al. (2003) found 350–570 mg (gallic acid equivalent)/100 g DW and Campos et al. (2012) determined a range of 790–3080 mg (chlorogenic acid equivalent)/100 g DW. The differences between these values and the lower ones registered in the present study can be a consequence of the treatment that the tubers went through to produce the flour, which means



**Fig. 3.** Total antioxidant capacity (ascorbic acid equivalent;  $n=9$ ) □ and total polyphenol content (gallic acid equivalent;  $n=6$ ) ■ of different weight<sub>yacon</sub>/volume<sub>solvent</sub> proportions, on a dry weight basis (extracts marked with the same letter, capitals for ascorbic acid equivalent values, are not significantly different ( $p > 0.05$ )) using water as solvent, boiling for 10 min.

that the processing of the yacon tubers lowered the total polyphenol content.

### 3.2. Phenolic composition LC–MS/MS

Analyses of the phenolic qualitative profile achieved by LC–MS/MS for the best yacon extract (Table 1) revealed the presence of four main phenolic compounds: chlorogenic acid, caffeic acid, coumaric acid and protocatechuic acid, as well as the amino acid tryptophan, which is in agreement with results reported by other research works (Campos et al., 2012; Simonovska et al., 2003; Yan et al., 1999). Relative quantification of phenolic compounds (fractional concentration percentage) showed that the most abundant compound was chlorogenic acid which represented 57.1% of the total peak area, followed by caffeic acid with 37.7%.

The methanolic extract (Table 1) revealed the presence of another phenolic compound in the flour, namely ferulic acid, which was however the most scarce (0.06% of the total peak area); this compound was also identified in yacon roots by Simonovska et al. (2003). In this methanolic extract the fractional concentration percentage of chlorogenic and caffeic acids were similar to that found in the aqueous extract.

Chlorogenic acid and caffeic acid have already been demonstrated to have antioxidant activity.

### 3.3. Biological assays

#### 3.3.1. DNA assay by agarose gel electrophoresis

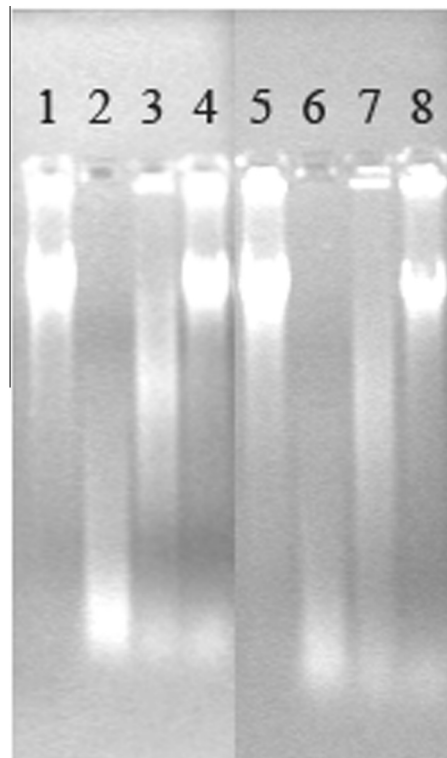
It is important to study the antioxidant activity via biological assays, since the antioxidant potential/properties of a certain extract can show very different results when a chemical method or a biological system is utilized. An extract possessing a high antioxidant capacity, as determined by a chemical method, is not necessarily a good biological antioxidant, and vice versa (Gião et al., 2010).

Results from the DNA assay showed that sterilized yacon tuber flour had an antioxidant effect, since when compared with the negative controls, i.e. oxidative degradation of  $H_2O_2$  alone (Fig. 4; lanes 2 and 6), it protected DNA in a large extent, since only a fraction of DNA was oxidized and degraded to small fragments (Fig. 4; lanes 3 and 7). Results also showed that sterilized yacon tuber flour does not have a pro-oxidant activity, since DNA did not show evident degradation when alone as observed in the positive controls (Fig. 4; lanes 1 and 5), assuring no pro-oxidant effect for the tested concentration (Fig. 4; lanes 4 and 8). Gião et al. (2008) also found artichoke and chamomile to show the same results (antioxidant and no pro-oxidant activities) upon DNA degradation. Another research work by Szeto, Chu, and Benzie (2006) makes use of a lysed cell version of the standard comet assay (in addition to the

**Table 1**  
Fractional concentration (%) of phenolic compounds, analyzed by LC–MS/MS.

Phenolic compound	M-1	Extract	
		Aqueous	Methanolic
Protocatechuic acid	153 > 109	0.23	0.22
Coumaric acid	163 > 119	3.63	4.38
Caffeic acid	179 > 135	37.68	40.63
Ferulic acid	193 > 134	–	0.06
Tryptophan	203 > 116	1.34	0.99
Chlorogenic acid	353 > 191	57.12	53.72
Total		100.00	100.00

Note: –, not detected.

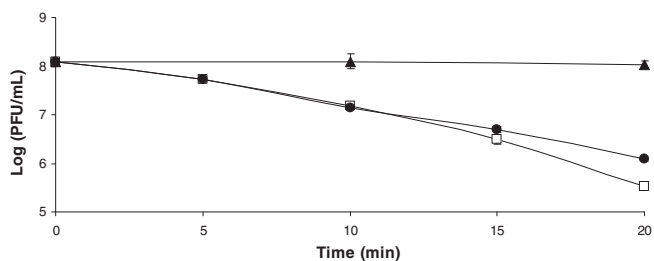


**Fig. 4.** Electrophoretogram from DNA assay for controls and sample: positive control – lanes 1 and 5; negative control – lanes 2 and 6; sample effect (in the presence of an oxidant–antioxidant effect) – lanes 3 and 7; and sample effect (in the absence of an oxidant – pro-oxidant effect) – lanes 4 and 8.

standard), in which “naked” DNA is exposed to putative genoprotectants or DNA-damaging agents, to study antioxidants in fruits and vegetables. Their results showed a protective effect of some fruits and vegetables upon “naked” DNA, but not all of them protected the whole cell in the standard comet assay. These results demonstrate the need to utilize more than one biological assay to determine the potential antioxidant activity upon DNA.

#### 3.3.2. Bacteriophages assay

The results obtained in this assay (Fig. 5) confirmed the results obtained from the DNA assay: a slight protection of phagic DNA oxidative degradation was observed (SOP), since the reduction in phages number was less than in the presence of  $H_2O_2$  alone (OP), thus exhibiting an antioxidant effect. This method also allowed to confirm the absence of pro-oxidant activity, because there was also no reduction in phages number when in contact with sterilized yacon tuber flour extract alone (SP). These results are similar to some of those obtained by Gião et al. (2010), when studying the antioxidant capacity of plant aqueous extracts: for aqueous



**Fig. 5.** Bacteriophage assay curves (SP – sample effect upon phage ▲; OP – oxidation of phage □; SOP – oxidation of phage in presence of sample ● (I); extent of antioxidant effect (calculated as the difference between the observed infection of the bacterium by the phage in the presence of sample and oxidant, and in the presence of oxidant only), throughout incubation time (II).

extracts such as raspberry and sage, after 20 min, a similar protection effect was observed.

This method is not to be extrapolated directly to eukaryotic cells; instead, it is intended for use as a complementary tool, to expand the study of the performance of antioxidant compounds *in vivo*. In terms of complexity, one might rank this system between a test on a DNA strand and a test on a whole eukaryotic cell (Gião et al., 2009).

Finally, both biological assays confirmed the relevant antioxidant capacity of the sterilized yacon tuber flour extract upon single DNA and phage DNA biological systems. They revealed that the extract had indeed antioxidant activity considering that it protected DNA from oxidative degradation, when directly exposed (DNA method) or enclosed in a phage (bacteriophage assay). Moreover, both methods proved that the extract at studied proportion of 8.9% (w/v) does not possess pro-oxidant activity, since it did not cause any damage of DNA or phage DNA when in contact with them.

#### 4. Conclusions

Sterilized yacon tuber flour showed relevant antioxidant properties, in particular due to its phenolic compounds composition. The antioxidant capacity of the extract determined by the ABTS<sup>+</sup> method was confirmed by both biological assays through direct protection of DNA and protection of phagic DNA, against oxidative degradation. These assays also showed the extract to not have pro-oxidant activity. A linear relationship between antioxidant activity and phenolics content was observed, and four main phenolic compounds were identified, where the two most abundant were chlorogenic acid followed by caffeic acid, two important antioxidants. The amino acid tryptophan which also has antioxidant activity was also identified in the extract.

Based on the results reported herein, i.e. the presence of antioxidant compounds (mainly phenolic compounds), and combined with scientific evidences of yacon tuber safety and health effects, as well as stability to food processing conditions, it can be stated that sterilized yacon tuber flour has the potential for being used by the food industry as an ingredient for the production of functional food products.

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