



Nutritional profile and anti-nutrient analyses of *Pachyrhizus ahipa* roots from different accessions



Cecilia Dini^a, María C. Doporto^a, María A. García^a, Sonia Z. Viña^{a,b,*}

^a CIDCA (Centro de Investigación y Desarrollo en Criotecología de Alimentos), Facultad Ciencias Exactas Universidad Nacional de La Plata (UNLP)–CONICET La Plata, 47 y 116 S/Nº, La Plata B1900AJJ, Buenos Aires, Argentina

^b Curso Bioquímica y Fitoquímica, Facultad Ciencias Agrarias y Forestales UNLP, Argentina

ARTICLE INFO

Article history:

Received 18 April 2013

Accepted 3 July 2013

Available online 13 July 2013

Keywords:

Leguminous tuberous roots

Starch and fiber

Radical scavenging activity

Phytic acid

Tripsin inhibitors

ABSTRACT

The food industry is interested in the study of sub-utilized plant species as a contribution to consumers' diet diversification and the incorporation of materials with specific technological properties. Thus, the chemical composition of *Pachyrhizus ahipa* roots belonging to different accessions was analyzed, focusing on components that show nutritional and technological relevance and including the analyses of the main anti-nutrients. Ahipa roots can be considered a good source of carbohydrates (sucrose and starch) and total dietary fiber (TDF). They can also supply a good quantity of minerals, mainly magnesium and iron. Protein levels are considerably higher than the ones reported for other root and tuber crops. Ahipa accessions Local and IRNAS 11 outstand for their high starch content (57 and 65% respectively), having a middle protein level (9–10.5%). Their TDF (21–22%) and sodium content were the lowest ones. Likewise, Local and IRNAS 11 accessions showed the lowest contents of oxalates, phytic acid and tannins, which represent an advantage from the nutritional point of view. Ahipa accessions exhibited differential solvent retention capacities and thermal stability. Local and IRNAS 11 accessions showed interesting properties to be considered in a breeding program since they had the highest starch content together with comparatively low content of anti-nutrients.

© 2013 Elsevier Ltd. All rights reserved.

1. Introduction

According to Opara (2003), root and tuber crops (R&T) encompass plants of diverse genera which are an important source of human carbohydrate food intake, mainly represented by the starch accumulated in their enlarged storage organs. These edible organs are from different botanical origin and structure and they include roots, tubers, corms, cormels, rhizomes, or bulbs.

Ahipa is the name that the Inca gave to the nutritious legume root produced by the American yam bean (*Pachyrhizus ahipa* (Wedd.) Parodi). The genus *Pachyrhizus* (Fabaceae) is native to Southern and Central America. Nowadays, *P. ahipa* is grown only in a few pockets of the Andean mountains of Bolivia and Peru, in the fertile valley floors at 1500–3000 m high and in the “ceja de selva” zone (N.R.C., 1989). It was also cultivated in Jujuy and Salta provinces in northern Argentina. The cultivation of *P. ahipa* is currently relegated (Leidi, Sarmiento, & Rodríguez Navarro, 2003). However, the plant is highly suited to the needs of small farmers since it has an efficient nitrogen

capturing root system and can be grown without nitrogen fertilizers input.

Ahipa protein content on a dry matter basis is higher than that of other R&T crops (N.R.C., 1989). Likewise, ahipa starch is easily digestible and the roots can be considered a good source of potassium. The crunchy roots are usually eaten raw in snacks and salads or it can also be boiled and accompany dishes in a similar way than cassava or sweet potato.

From the industrial point of view, the main interest in *P. ahipa* is the starch extraction. This crop can be considered an alternative starch source to other R&T produced for this purpose. In previous works, some relevant technological properties of the starch and flours derived from *P. ahipa* were studied (Doporto, Dini, Mugridge, Viña, & García, 2012; Doporto, Mugridge, García, & Viña, 2011; López et al., 2010). However, the chemical composition as well as the nutritional and technological properties of ahipa roots belonging to different accessions has not been fully investigated.

Among the minor components of plant tissues, some of them are mentioned as anti-nutrients. According to Novak and Haslberger (2000), some discrepancy persists about the definition of plant inherent toxicants and anti-nutrients. Anti-nutrients are usually understood as substances that inhibit or block important pathways in the metabolism, especially the digestion. They also reduce the maximum utilization of nutrients and interfere with an optimal assimilation of the proteins, vitamins or minerals present in a food, decreasing its nutritive value.

* Corresponding author at: CIDCA (Centro de Investigación y Desarrollo en Criotecología de Alimentos), Facultad Ciencias Exactas Universidad Nacional de La Plata (UNLP)–CONICET La Plata, 47 y 116 S/Nº, La Plata (B1900AJJ), Buenos Aires, Argentina. Tel.: +54 221 424 9287; fax: +54 221 425 4853.

E-mail address: soniavia@quimica.unlp.edu.ar (S.Z. Viña).

Most anti-nutrients are plant secondary metabolites, which act as natural chemical defense mechanisms, protecting plants from herbivores and pathogenic microorganisms (Dini, Garcia, & Viña, 2012). They are usually consumed in small amounts to cause severe or lethal effects, but in some specific cases they can lead to life-threatening toxic reactions (Dini et al., 2012). The works referred to the content of anti-nutrients present in *P. ahipa* roots are rather scarce, particularly those studies concerning different ahipa accessions.

The objective of the present work was to analyze the chemical composition of *P. ahipa* roots belonging to different accessions, focusing on those components that show nutritional and technological relevance and including the analyses of the main groups of anti-nutrients.

2. Materials and methods

2.1. Plant material

P. ahipa plants were grown at the EEA-INTA Montecarlo farm (Misiones, Argentina). The accessions Local (originally from Perico, Jujuy, Argentina) and IRNAS N° 4, 5, 9 and 11 (originally from Las Cabezas, Sevilla, Spain) were studied. The lines used in this work were selected according to their agronomic performance. Roots were harvested in April 2010. They were received at the laboratory and processed immediately. Wounded and unhealthy roots were removed. Selected roots were thoroughly washed with tap water and they were sanitized by immersion in NaClO solution (250 ppm of chlorine, 10 min). Subsequently, roots were dried at room temperature, sliced and dried in an oven at 60 °C until constant weight was achieved. They were grinded and kept into hermetically closed jars until analyses were performed. This material corresponds to “partially dehydrated roots”.

2.2. Chemical composition of ahipa roots

2.2.1. Dry matter and total ash content

Residual water was gravimetrically quantified by placing ahipa roots samples (0.5 g) in an oven (San Jor H701P, Argentina) at 105 °C until reaching constant weight. The results were expressed as percentage (%) of the initial weight. Total ash quantification was also performed gravimetrically after incineration in a muffle furnace (Indef 331, Córdoba, Argentina) at 550 °C. The percentage (%) of total ash on a dry basis was calculated.

2.2.2. Sodium, potassium, calcium, magnesium and iron content

The mineral analyses were performed by atomic absorption or emission spectroscopy following the methods described by Dopporto et al. (2011). The final results were expressed as percentage (%) on a dry basis.

2.2.3. Liposoluble fraction

Partially dehydrated roots (12 g) were extracted with hexane in a Soxhlet apparatus until completing eight cycles of extraction. The residue obtained in the flask after solvent recovery and evaporation (total fat) was weighed. The results were expressed as percentage (%) on a dry basis.

2.2.4. Crude protein

Partially dehydrated roots (0.8 g) were analyzed for total nitrogen content by the Kjeldahl method (AOAC, 1990). The results were expressed as percentage (%) on a dry basis.

2.2.5. Total starch (TS) and total dietary fiber (TDF)

The content of TS and TDF in partially dehydrated ahipa roots samples were measured by the enzymatic kits K-TSTA 05/06 and K-TDFR 05/12 Megazyme® (Ireland) respectively. The results were expressed as percentage (%) on a dry basis.

2.2.6. Sugar content

Ahipa roots from different accessions were analyzed for their glucose, fructose and sucrose content by the enzymatic kit K-SUFRG 12/05 Megazyme® (Ireland). Saccharides were extracted with distilled water and the results were expressed as percentage (%) on a dry basis.

2.2.7. Radical scavenging activity (RSA)

The analysis of the RSA of ahipa roots was performed with the 2,2-diphenyl-1-picrylhydrazyl (DPPH•) stable radical, as reported by Brand-Williams, Cuvelier, and Berset (1995), and with the radical monocation of 2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS•⁺) following the protocol of Re et al. (1999). In both cases, determinations were carried out on fresh frozen root tissues from the IRNAS 11 and Local accessions, which were grinded and extracted with ethanol 96%. The amount of frozen tissue needed to decrease by 50% the initial DPPH• concentration was calculated and defined as EC₅₀. Results were expressed as EC₅₀⁻¹ (g⁻¹). For RSA evaluation with ABTS•⁺, Trolox was used as a standard (0–0.432 mg mL⁻¹) and the results were expressed as equivalent mg Trolox/100 g sample.

2.2.8. Anti-nutrient analyses

Phytate was extracted with HCl 3.5% and quantified following the protocol described by Makkar, Siddhuraju, and Becker (2007). Separation was performed in an anionic exchange column (CG-400 Type I chloride-saturated resin, 100–200 mesh, Rohm & Haas, Philadelphia B, PA.) with 0.8 cm diameter and 9.5 cm height. Phytate quantification was performed by the reaction with Wade's Reagent (Vaintraub & Lapeva, 1988). Phytic acid was used as standard. Phytate contents were expressed as mg phytic acid/g sample. Oxalate determination was performed by extraction of grinded sample with 0.3 M HCl and was quantified according to AOAC (1990). Results were expressed as mg oxalate/100 g. Trypsin inhibition activity was calculated as milligrams of pure trypsin inhibited per g of sample (mg/g) extracted with 0.01 M NaOH following the protocol described in Makkar et al. (2007); based on Kakade, Simons, and Liener (1969) and Smith, Van Megen, Twaalfhoven, and Hitchcock (1980). Tannins and total phenolics were extracted using aqueous acetone (70%) and the quantification was performed using the Folin–Ciocalteu method described in Makkar et al. (2007), based on Makkar, Blümmel, Borowy, and Becker (1993). The results were expressed as mg tannic acid/g.

Likewise, the presence of cyanogenic glycosides was qualitatively investigated from macerated ahipa root samples by the reduction of sodium picrate to a red colored compound (Makkar et al., 2007).

2.3. Properties related to technological aspects

Solvent retention capacity (SRC) of partially dehydrated ahipa root samples were analyzed following the AACC method 56-11.02 (Rosell, Santos, & Collar, 2009). The solvents used were deionized water, sucrose (50% w/w), Na₂CO₃ (5% w/w) and lactic acid (5% w/w) solutions. SRC was calculated as follows and expressed as percentage (%):

$$\% \text{ SRC} = \left[\frac{\text{Gelweight}}{\text{Sampleweight}} - 1 \right] \times \left[\frac{86}{(100 - \% \text{ Samplewatercontent})} \right] \times 100. \quad (1)$$

Thermal properties of partially dehydrated ahipa root samples or the derived flours were determined according to Dopporto et al. (2011) using a Q100 DSC controlled by a TA 5000 module (TA Instruments, New Castle, Delaware, USA) with a quench-cooling accessory, under a N₂ atmosphere (20 mL/min). For these analyses, ahipa flours were obtained as described in a previous work (Dopporto et al., 2011).

2.4. Statistical analysis

A completely randomized experimental design was used. The whole experiments were performed in duplicate, being the replicated experimental units a pool of at least 10 roots. Most analytical determinations were carried out at least by triplicate. Analysis of variance (ANOVA) and comparison of means with the Fisher's least significant difference (LSD) test were conducted, at a significance level $p = 0.05$.

3. Results and discussion

3.1. Chemical composition of ahipa accessions

Moisture content of ahipa roots was 78.4–83.5% (Table 1), similar to the values reported by Ørting et al. (1996) and Leonel, Sarmiento, Cereda, and Cãmara (2003). These moisture levels are similar to those found in potato (*Solanum tuberosum*, 76–79%), and higher than the ones reported for cassava (*Manihot esculenta*, 63–65%) (Dini et al., 2012).

Ahipa total ash content was in the range 3.32–4.20% (Table 1). These levels were higher than the ones reported for other R&T crops such as yam (*Dioscorea alata*) (0.88–0.91%), taro (*Colocasia esculenta*) (0.73–1.04%), cassava (*M. esculenta*) (1.15%) (Leterme, Buldgen, Estrada, & Londoño, 2006) and sweet potato (*Ipomoea batatas*) (2.15–2.95%) (Ahmed, Sorifa, & Eun, 2010). Ahipa total ash content was similar to that reported for yacon (*Smallanthus sonchifolius*) according to Choque Delgado, da Silva Cunha Tamashiro, Maróstica Junior, and Pastore (2013). Ahipa accession IRNAS 5 outstands for its high total ash content.

Accessions Local and IRNAS 11 had higher starch content than the others (Table 1). Cassava is richer than ahipa in starch content. However, ahipa showed good starch extraction yield (56.5% dry basis) (López et al., 2010). An agronomic advantage of ahipa crop propagation is that it can be performed from the seeds instead of the roots, unlike it occurs with cassava. This characteristic implies that a greater proportion of roots can be destined to the starch extraction process in an industrial scale.

From a nutritional point of view, the most relevant feature of *P. ahipa* compared to other R&T is its relatively high protein content. Accessions Local and IRNAS 11 had a statistically ($p > 0.05$) similar protein content and this level was higher than that of the accession IRNAS 9, but lower than the one found in IRNAS 4 (Table 1). These protein values are considerably higher than those found in other R&T such as cassava (1–2%), potatoes (3–6%), sweet potato (1–10%), taro (0.6–2%), yams (1–3%) (Shewry, 2003) or yacon (2.45–6.48%) (Choque Delgado et al., 2013). Although ahipa is a rich source of protein, the study of the amino acids profile of ahipa proteins becomes critical, in order to better assess its nutritional quality.

Crude fat levels were within the range 0.43–0.63% (Table 1). This low lipid content, which was similar to that reported for sweet potato (Ahmed et al., 2010) and yacon (Choque Delgado et al., 2013), positions ahipa as a possible ingredient for the production of low-fat foods.

Among the macrocomponents, total dietary fiber (TDF) was also quantified. Dietary fiber is the collective name given to all components of plant cell walls which are not hydrolysed by enzymes in the human

digestive tract. TDF has been recognized to produce beneficial effects on the intestinal function regulation, preventing colonic disorders and diverticula (Steinmetz & Potter, 1996). Ahipa roots contained TDF within the range 20.8–25.9 g/100 g (dry basis). Roots from the accessions IRNAS 11 and Local showed the lowest content (Table 1). Accessions IRNAS 5 and 9 outstand for its high TDF levels. These results show that ahipa roots can be considered a good source of TDF compared to other R&T. For example, Castro, Céspedes, Carballo, Bergenstahl, and Tornberg (2013) reported 10.4 g TDF/100 g in yacon. Taking into account that the dietary fiber intake should range 20–35 g/day for optimal benefits (Dreher, 2001) 100 g of dehydrated ahipa roots almost supply the daily requirement for fiber.

Table 2 shows the mineral content of ahipa roots. In general, mineral content varied significantly with ahipa accession. The samples from accession IRNAS 5 exhibited the highest Na content while those from accessions Local and IRNAS 11 presented the lowest values.

Sodium concentration in ahipa was higher than in cassava (36–50 mg/100 g) (Charles, Sriroth, & Huang, 2005) or taro (41.5 to 120.9 mg/100 g) (Lewu, Adebola, & Afolayan, 2010). This is important because it is widely recognized that a high intake of sodium is related to the onset of hypertension, increasing the risk of heart attacks, strokes and kidney damage (Dini et al., 2012; Havas, Roccella, & Lenfant, 2004). Taking this consideration into account, accession IRNAS 5 roots would not be appropriate for low sodium product formulations.

Ahipa calcium content was similar to that reported for yacon roots (Choque Delgado et al., 2013).

The values of Mg found in ahipa (Table 2) were higher than those reported for cassava by Charles et al. (2005) (31–43 mg/100 g). It is worth noting that an insufficient Mg intake has been associated with the development of diabetes and insulin resistance as well as cardiovascular complications (Ascherio et al., 1998; Saris, Mervaala, Karppanen, Khawaja, & Lewenstam, 2000).

Daily recommended iron intake is in the range of 25–50 mg (Gurzau, Neagu, & Gurzau, 2003). The contribution of Fe from ahipa (Table 2) varies with the accession. The accession IRNAS 5 exhibited the highest value respect to the others. The content of this mineral in the accession IRNAS 4 (630 mg/100 g dry matter) was similar to that of sweet potato (Eluagu & Onimawo, 2010). The contents of the other accessions (126–173 mg/100 g) were considerably higher compared with other R&T such as cassava (29–40 mg/100 g) (Charles et al., 2005) or taro, whose values were below the detection limit of the technique in seven accessions analyzed (Lewu et al., 2010).

Likewise, ahipa samples contained high sugar concentrations when comparing with other R&T. In communities where the cultivation and marketing of *P. ahipa* is a common practice, the root is mainly consumed raw, since it resembles a fruit due to its high water content and sweet taste. Table 3 shows the sugar content of ahipa roots. Ahipa accession IRNAS 4 presented the highest values of sucrose while IRNAS 11 exhibited the lowest levels of glucose, fructose and sucrose. The average values found for simple sugars in all the tested accessions were between 14.1–25.1 g/100 g on dry basis. These values are close to those reported for sweet potato: 17.17–20.76% (Ahmed et al., 2010), a product which also has a characteristic of sweet taste. When comparing with other

Table 1
Chemical composition (% w/w) of *Pachyrhizus ahipa* roots from different accessions.

Accession	Dry matter content (quantified at 60 °C)	Dry matter content (quantified at 105 °C)	Total ash	Crude fat	Crude protein	Total starch content	Total dietary fiber
Local	21.6 ± 0.3 ^a	87.0 ± 0.3 ^a	3.32 ± 0.07 ^d	0.48 ± 0.01 ^b	10.5 ± 0.5 ^{a,b}	56.8 ± 0.9 ^{a,b}	22.4 ± 0.6 ^c
IRNAS N° 4	18.3 ± 1.2 ^b	82.9 ± 0.3 ^c	3.82 ± 0.00 ^b	0.43 ± 0.07 ^b	11.5 ± 1.2 ^a	53.9 ± 6.2 ^b	24.4 ± 0.8 ^b
IRNAS N° 5	16.5 ± 1.5 ^b	85.8 ± 0.5 ^b	4.20 ± 0.04 ^a	0.63 ± 0.03 ^a	9.6 ± 0.3 ^b	54.1 ± 3.2 ^b	25.2 ± 0.5 ^{a,b}
IRNAS N° 9	18.2 ± 0.9 ^b	85.1 ± 0.1 ^b	3.81 ± 0.02 ^{b,c}	0.62 ± 0.00 ^a	7.9 ± 0.5 ^c	43.7 ± 1.6 ^c	25.9 ± 0.2 ^a
IRNAS N° 11	20.8 ± 0.9 ^a	87.3 ± 0.3 ^a	3.70 ± 0.05 ^c	0.47 ± 0.02 ^b	9.0 ± 0.3 ^{b,c}	65.0 ± 0.6 ^a	20.8 ± 0.2 ^d

Note: Reported values correspond to the mean ± standard deviation. Results are expressed on dry basis. Different letters in the same column indicate significant differences ($p < 0.05$).

Table 2
Mineral content (mg/100 g) of *Pachyrhizus ahipa* roots from different accessions.

Accession	Sodium	Potassium	Calcium	Magnesium	Iron
Local	754 ± 8 ^d	129 ± 4 ^c	415 ± 7 ^d	531 ± 4 ^c	173 ± 6 ^c
IRNAS N° 4	917 ± 9 ^c	172 ± 4 ^b	573 ± 8 ^b	164 ± 4 ^e	630 ± 7 ^b
IRNAS N° 5	1302 ± 10 ^a	172 ± 5 ^b	546 ± 9 ^c	227 ± 5 ^d	756 ± 8 ^a
IRNAS N° 9	945 ± 9 ^b	168 ± 4 ^b	815 ± 8 ^a	785 ± 4 ^a	168 ± 7 ^c
IRNAS N° 11	207 ± 9 ^e	192 ± 4 ^a	181 ± 8 ^e	592 ± 4 ^b	126 ± 7 ^d

Note: Reported values correspond to the mean ± standard deviation. Results are expressed on dry basis. Different letters in the same column indicate significant differences ($p < 0.05$).

Andean crops such as maca (*Lepidium meyenii*), values are similar too (17.57% of total sugars in maca roots), according to Rondán-Sanabria, Valcarcel-Yamani, and Finardi-Filho (2012). Among the sugars present in ahipa roots, sucrose was predominant (Table 3).

3.2. RSA and anti-nutrients factors

Ahipa Local accession showed significantly higher RSA (42.5 ± 0.7 mg Trolox/100 g) measured by the ABTS⁺ method than the accession IRNAS 11 (28.6 ± 0.9 mg Trolox/100 g); these values were similar to that information for sweet potato (25.03 ± 4.07 mg Trolox/100 g) but lower than those of cassava (51.07 ± 7.10 mg Trolox/100 g) or the ordinary yam (*Typhonium trilobatum*) (74.05 ± 10.20 mg Trolox/100 g) (Sreeramulu & Raghunath, 2010).

DPPH• scavenging assays showed that the inactivation reaction took 6 h to reach the steady state, so ahipa root antioxidants could be included into the group with slow reaction kinetics (Brand-Williams et al., 1995). This would indicate a relatively low quantity of ascorbic acid as well as a major contribution of phenolics to the antioxidant power. Several phenolic compounds such as coumaric, ferulic, caffeic and gallic acid among others, showed slow kinetic behavior for the inactivation of DPPH• meanwhile ascorbic and isoascorbic acid brought about a rapid kinetic behavior (Brand-Williams et al., 1995). The RSA exhibited by ahipa roots against DPPH• was 2.73 ± 0.36 g⁻¹ (EC₅₀¹) for accession IRNAS 11 and 2.51 ± 0.37 g⁻¹ for the Local one. Despite the antioxidant levels of ahipa roots did not differ from those reported for other R&T, the incorporation of this tuberous root into the diet may not represent a significant antioxidant input, since other vegetables can provide markedly more antioxidants than ahipa roots.

In the analysis of anti-nutrients factors, phytin was detected in all the analyzed accessions. Values presented in Table 4 show phytate contents ranging from 1.0 to 1.7 mg phytic acid/g sample, similar to those reported for cassava flour (0.9–1.4 mg/g) (Charles et al., 2005) and potato (1.1–2.7 mg/g) (Phillippy, Lin, & Rasco, 2004) but lower than the values found in legumes such as soybean (12.0–17.5 mg/g) and peas (7.2–12.3 mg/g) (Hidvegi & Lasztity, 2002) and cereals such as wheat (9.6–22.2 mg/g); maize (10.8 mg/g); rye (4.5–5.7 mg/g) and rice (5.5–13.5 mg/g) (García-Esteva, Guerra-Hernández, & García-Villanova, 1999; Kikunaga, Takahashi, & Huzisige, 1985; Wu, Tian, Walker, & Wang, 2009). Phytate, released from phytic acid dissociation, is typically related to a reduction in iron biodisponibility.

Table 3
Simple sugar content (g/100 g) of *Pachyrhizus ahipa* roots from different accessions.

Accession	Glucose	Fructose	Sucrose
Local	1.67 ± 0.15 ^b	5.41 ± 0.25 ^b	11.44 ± 0.07 ^b
IRNAS N° 4	1.77 ± 0.07 ^b	6.29 ± 0.07 ^a	17.01 ± 0.42 ^a
IRNAS N° 5	1.57 ± 0.10 ^b	5.34 ± 0.03 ^b	7.82 ± 0.18 ^d
IRNAS N° 9	2.81 ± 0.10 ^a	6.33 ± 0.16 ^a	10.23 ± 0.39 ^c
IRNAS N° 11	0.74 ± 0.03 ^c	4.51 ± 0.01 ^c	8.89 ± 0.13 ^d

Note: Reported values correspond to the mean ± standard deviation. Results are expressed on dry basis. Different letters in the same column indicate significant differences ($p < 0.05$).

Besides iron, phytate can also precipitate magnesium, zinc, copper, calcium and manganese and it is classified within the antimicrobial compounds. Contrary to the adverse effects described, phytate has been proven to have anticarcinogenic activity on K-562 and H-29 cells and on laboratory animals (rats and mice) (Harland & Morris, 1995) as well as an antioxidant capacity (Rimbach & Pallauf, 1998). A linear correlation ($r^2 > 0.82$) was found between calcium and phytate content in partially dehydrated ahipa roots.

Concerning the quantification of phenolic compounds (Table 4), total phenolic content in IRNAS 11 accession was significantly lower ($p < 0.05$) than the one measured in IRNAS 4, 5, 9 and did not differ ($p > 0.05$) from that registered for the Local accession. When comparing to other R&T crops, reported values for total phenolic content (expressed as gallic acid equivalents mg/100 g) are: 81.59 ± 21.03 for colocasia (*Colocasia antiquorum*); 38.42 ± 0.62 for potato (*S. tuberosum*); 53.70 ± 3.44 for sweet potato (*I. batatas*); 137.55 ± 6.04 for cassava (*M. esculenta*) and 54.92 ± 8.15 for ordinary yam (*T. trilobatum*) (Sreeramulu & Raghunath, 2010). Ahipa accessions had tannin values between 1.6 and 2.0 mg of tannic acid/g sample, which were comparable to those reported in cassava flour (1–2 mg/g) (Obboh & Akindahunsi, 2003) and sweet potato (2.5 mg/g) (Eluagu & Onimawo, 2010), but lower than those normally present in bean (Guzmán-Maldonado, Castellanos, & De Mejía, 1996). The presence of tannins becomes relevant since they act as inhibitors of digestive enzymes and can also reduce the palatability of a food for producing astringency when combined with the salivary proteins. On the other hand, certain types of tannins have been mentioned as reducing the risk of coronary heart disease (Dell'Agli, Busciana, & Bosio, 2004; Gresele et al., 2011). Tannins represented approximately 50% of the total phenolic compounds in the analysis of ahipa accessions (Table 4).

Trypsin inhibitory activity of ahipa roots, expressed as trypsin inhibitor units (TIU), was found between 1.0 and 2.7 TIU/mg (Table 4), comparable to the values reported for legumes with lowest trypsin inhibitor activity such as chickpeas, lentils and lupins, and cereals such as maize, which are generally between 1 and 20 TIU/mg. Soybeans may have concentrations above 40 TIU/mg (Dini et al., 2012). Since assays were carried out on partially dehydrated roots, it is important to stress that the remaining activities of trypsin inhibitors were determined on ahipa samples. Part of the total trypsin inhibition activity could have been inactivated by the heating process. The Local and IRNAS 11 accessions exhibited the highest values while those from accessions IRNAS 4, 5 and 9 presented similar and lower trypsin inhibitor contents.

Trypsin inhibitors can cause pancreatic hypertrophy and/or hyperplasia (Makkar et al., 2007). However, it has been reported that protease inhibitors have anticarcinogenic effects (Dragsted, Strube, & Larsen, 1993; Schelp & Pongpaew, 1988; Steinmetz & Potter, 1996). Likewise, in a controlled consumption, antiprotein activity of these substances can be used to produce satiety in foods designed for weight reduction.

The oxalate content of the ahipa roots varied between 58 and 217 mg/100 g (Table 4). Huang, Chen, and Wang (2007) reported an oxalate content of 234–411 mg/100 g in taro. Roots from accessions IRNAS 4 and 9 showed statistically ($p > 0.05$) similar and higher oxalate contents while those from the accessions Local and 11 showed the lowest values and no significant differences ($p > 0.05$) were found between them. Oxalates reduce the bioavailability of calcium, and the insoluble complexes formed can also produce blockage of the renal tubules and development of urinary calculi (Dini et al., 2012; Hang, Vanhanen, & Savage, 2013; Sasaki et al., 2008). A linear correlation ($r^2 > 0.75$) was found between calcium and oxalate content in ahipa roots.

Interestingly, ahipa roots from Local and IRNAS 11 accessions presented the lowest contents of oxalates, phytic acid and tannin within the ranges observed. Likewise, as stated in Materials and methods section, the presence of cyanogenic glycosides was qualitatively investigated from macerated ahipa root samples and the levels were not detectable.

Table 4Anti-nutrient components and total phenolic compounds of partially dehydrated *Pachyrhizus ahipa* roots from different accessions.

Accession	Tannins (mg tannic acid/g)	Phytic acid (mg/g)	Tripsin inhibition activity (trypsin inhibitors units TIU/mg)	Oxalates (mg/100 g)	Total phenolic compounds (mg/g)
Local	1.70 ± 0.06 ^b	1.03 ± 0.01 ^b	2.73 ± 0.02 ^a	74.34 ± 7.51 ^c	3.21 ± 0.36 ^b
IRNAS N° 4	1.92 ± 0.02 ^a	1.27 ± 0.00 ^b	1.05 ± 0.03 ^c	217.39 ± 7.50 ^a	3.77 ± 0.22 ^a
IRNAS N° 5	2.03 ± 0.05 ^a	1.33 ± 0.27 ^b	1.02 ± 0.01 ^c	176.43 ± 5.63 ^b	3.79 ± 0.30 ^a
IRNAS N° 9	1.94 ± 0.00 ^a	1.73 ± 0.02 ^a	1.01 ± 0.10 ^c	209.86 ± 3.76 ^a	3.73 ± 0.06 ^a
IRNAS N° 11	1.59 ± 0.01 ^b	1.05 ± 0.02 ^b	2.26 ± 0.12 ^b	58.37 ± 7.50 ^c	3.03 ± 0.06 ^b

Note: Reported values correspond to the mean ± standard deviation. Different letters in the same column indicate significant differences ($p < 0.05$).

3.3. Solvent retention capacity (SRC)

Fig. 1 shows SRC of partially dehydrated roots from different ahipa accessions. In general, accession IRNAS 4 exhibited the highest retention capacity for the solvents tested while accession IRNAS 11 presented the lowest values.

Sodium Carbonate SRC (SCRC) is believed to be related to the damaged starch content of the flour, since a 5% (w/w) sodium carbonate solution elevates the pH above 11 (Gaines, 2000) where starch hydroxyl groups begin to ionize and starch is hence negatively charged (Duyvejonck, Lagrain, Pareyt, Courtin, & Delcour, 2011). Ahipa roots from accessions IRNAS 9 and 5 could contain a similar content of damaged starch since their SCRC did not differ significantly ($p > 0.05$) (Fig. 1).

In general, Lactic Acid SRC (LARC) is associated with glutenin network formation and the gluten strength of flour, because a pH below 7 favors swelling and network formation by gluten polymers relative to polysaccharides (Duyvejonck et al., 2011). Since ahipa is a gluten free product, LARC can be associated to the protein capacity to develop a network. Thus, although IRNAS 11 and Local accessions exhibited higher protein content quantified by the Kjeldahl method (Table 1) they would not be suitable for protein network formation. This is of primary importance in the development of products for celiac population, since dough characteristics of ahipa-based products should be enhanced by the incorporation of hydrocolloids and specific additives.

A strong linear correlation ($r^2 > 0.84$) between TDF and water retention capacity (WRC) was observed. A similar trend was reported for wheat flour (Colombo, Pérez, Ribotta, & León, 2008; Duyvejonck, Lagrain, Dornez, Delcour, & Courtin, 2012; Duyvejonck et al., 2011).

Partially dehydrated roots from IRNAS 11 and Local accessions showed lower sucrose retention capacity (SuRC), which is associated to water-soluble pentosan content, quantified as soluble fiber. These findings suggest that these accessions would contain higher proportions of insoluble fiber with the health benefits that this entails. Besides, SuRC also correlated with TDF ($r^2 > 0.77$).

The SRC tests are commonly used to establish flour quality and practical functionality profile which is useful for predicting its performance in specific baking applications. Thus, both partially dehydrated

root and flour SRC assays were conducted. The results showed that SRC of partially dehydrated roots were higher than those of ahipa flours from different accessions. The comparison is shown in Fig. 2 for Local ahipa accession as an example. Since partially dehydrated samples include the root peel which makes an important contribution of TDF, their WRC was significantly ($p < 0.05$) higher than the corresponding flour. A similar trend was observed for SCRC, indicating a lower damaged starch in the flour. The higher SuRC in partially dehydrated roots would suggest a higher proportion of components related to soluble dietary fiber. With regard to LARC, the flour obtained by the whole ahipa root processing would lead to enhance its baking quality since the development of the protein network would be favored, which constitutes a technological improvement.

3.4. Thermal properties

In general, partially dehydrated ahipa root aqueous suspensions exhibited a wide transition mainly associated to starch gelatinization and protein denaturalization, being both cooperative processes (Fig. 3). The Table inset in Fig. 3 shows that a similar trend was followed by the onset peak temperature. Partially dehydrated ahipa roots from accessions Local and IRNAS 11 did not differ statistically ($p > 0.05$) while differences between other accessions were significant ($p < 0.05$). Accessions Local and IRNAS 11, with high starch and protein contents (Table 1), exhibited the highest enthalpy values. Likewise, accession IRNAS 4 with the highest sucrose content (Table 3) presented the highest peak temperature; it is well known that sugars shift the peak temperature of starch gelatinization and also affect the protein denaturalization process due to water availability reduction.

Fig. 4 compares the thermal behavior of partially dehydrated roots and the derived flour for Local ahipa accession; a similar trend was observed in the onset peak temperature as well as the enthalpy associated. Likewise, the thermal parameters of ahipa flour and starch from Local ahipa accession were previously studied (Dopporto et al., 2011). Peak temperature corresponding to flour was higher than that of the derived starch (Table inset in Fig. 4). A similar trend was

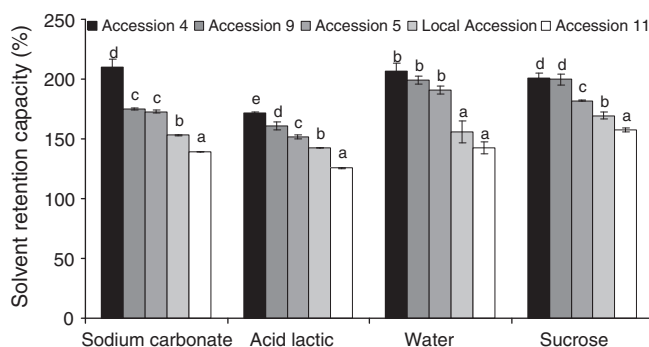


Fig. 1. Solvent retention capacity of *P. ahipa* partially dehydrated roots from different accessions.

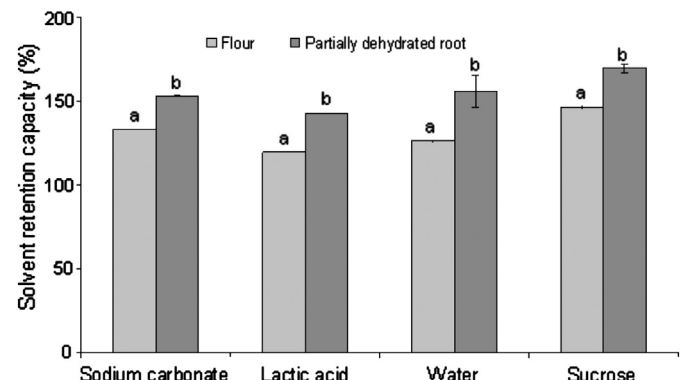
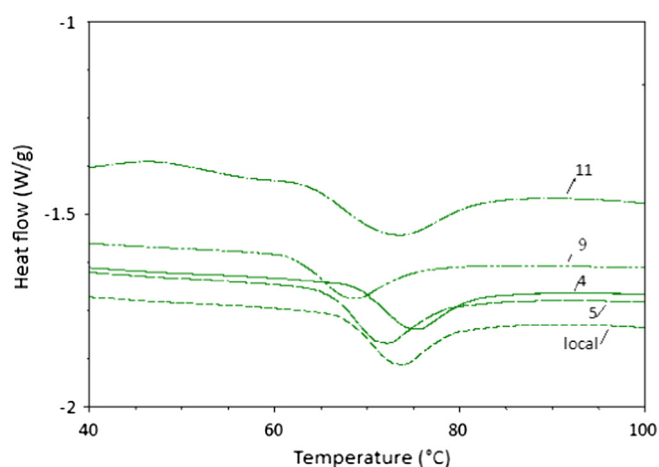


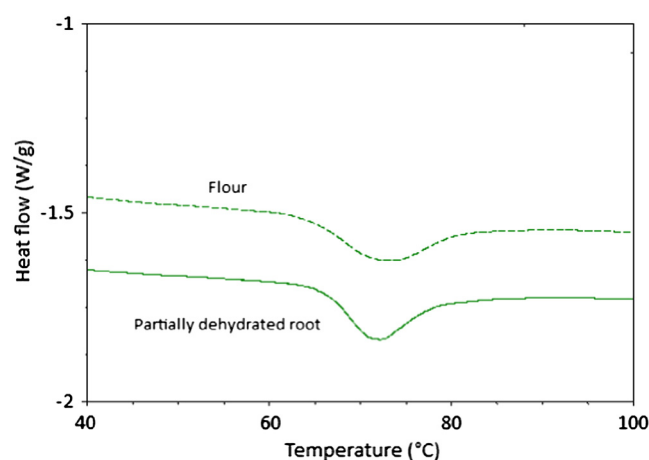
Fig. 2. Solvent retention capacity of *P. ahipa* partially dehydrated roots and flour from Local accession.



Accession	Onset peak temperature (°C)	Peak temperature (°C)	Enthalpy (J / g dry basis)
IRNAS 4	69.7 ± 0.7 ^d	75.3 ± 0.5 ^d	17.5 ± 0.5 ^a
IRNAS 5	68.0 ± 0.4 ^c	73.5 ± 0.2 ^c	22.6 ± 3.2 ^b
IRNAS 9	62.2 ± 0.4 ^a	68.2 ± 0.2 ^a	20.6 ± 0.4 ^b
IRNAS 11	66.1 ± 0.4 ^b	71.9 ± 0.5 ^b	25.4 ± 0.9 ^c
Local	66.1 ± 0.1 ^b	71.3 ± 0.4 ^b	24.7 ± 2.9 ^c

Reported values correspond to the mean ± standard deviation. Results are expressed on dry basis. Different letters in the same column indicate significant differences ($p < 0.05$).

Fig. 3. DSC thermograms and thermal parameters of *P. ahipa* partially dehydrated roots from different accessions.



Sample	Onset peak temperature (°C)	Peak temperature (°C)	Enthalpy (J / g dry basis)
Partially dehydrated root	66.1 ± 0.1 ^b	71.3 ± 0.4 ^b	24.7 ± 2.9 ^b
Flour	64.5 ± 0.1 ^a	72.0 ± 0.3 ^b	8.2 ± 0.6 ^a
Starch	64.3 ± 0.2 ^a	67.2 ± 0.1 ^a	9.6 ± 0.6 ^a

Reported values correspond to the mean ± standard deviation. Results are expressed on dry basis. Different letters in the same column indicate significant differences ($p < 0.05$).

Fig. 4. DSC thermograms and thermal parameters of *P. ahipa* partially dehydrated roots and flour from Local accession.

observed in other works performed on cassava and ahipa (Aboubakar, Njintang, Scher, & Mbofung, 2008; Doporto et al., 2011, 2012; López et al., 2010).

4. Conclusions

Several processed food products obtained from *Pachyrhizus* roots have been mentioned, such as flour, stirred yogurt with dietary fiber, juice, fermented beverages, etc. (Ramos-de-la-Peña, Renard, Wicker, & Contreras-Esquivel, 2013) that evidences a growing perspective of use. Thus, the characterization of ahipa accessions will provide useful information for the development of food derivatives and ingredients.

The chemical composition of ahipa roots varied with the accession of origin. Although ahipa water content is relatively high, the roots can be considered a good source of carbohydrates (sucrose and starch) and TDF. They can also supply a good quantity of minerals, mainly magnesium and iron. Protein levels are considerably higher than the ones reported by other R&T crops. However, future research demands the analyses of the aminoacid profile of ahipa protein. Ahipa accessions Local and IRNAS 11 outstand for their high starch content, presenting a middle protein level. The TDF and Na content of both accessions were the lowest ones. Likewise, Local and IRNAS 11 accessions showed the lowest contents of oxalates, phytic acid and tannins, which represent an advantage from the nutritional point of view. IRNAS 11 had the lowest values of SRC and the LARC values in particular would indicate that proteins present in these materials would not be suitable for protein network formation. This is a technological aspect that should be improved when designing raw materials and ingredients for food production for celiac patients from ahipa roots. When analyzing thermal properties, the accessions Local and IRNAS 11 exhibited the highest enthalpy values. Thus, both materials showed interesting properties to be considered in a breeding program.

Acknowledgments

This work was financially supported by the Projects PICT 2007-1100 and PICT 2011-1213 (ANPCyT). The authors wish to thank INTA Montecarlo for providing ahipa roots and Mrs Alicia Mugridge (CIDCA) for her assistance in roots processing and preparation.

References

- Aboubakar, Njintang, Y. N., Scher, J., & Mbofung, C. M. F. (2008). Physicochemical, thermal properties and microstructure of six varieties of taro (*Colocasia esculenta* L. Schott) flours and starches. *Journal of Food Engineering*, 86(2), 294–305.
- Ahmed, M., Sorifa, A. M., & Eun, J. B. (2010). Effect of pretreatments and drying temperatures on sweet potato flour. *International Journal of Food Science and Technology*, 45(4), 726–732.
- AOAC (1990). In K. Helrich (Ed.), *Official methods of analysis of the Association of Official Analytical Chemists* (15th ed.). Arlington, Va: Association of Official Analytical Chemists.
- Ascherio, A., Rimm, E. B., Hernán, M. A., Giovannucci, E. L., Kawachi, I., Stampfer, M. J., et al. (1998). Intake of potassium, magnesium, calcium, and fiber and risk of stroke among US men. *Circulation*, 98, 1198–1204.
- Brand-Williams, W., Cuvelier, M. E., & Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. *LWT – Food Science and Technology*, 28(1), 25–30.
- Castro, A., Céspedes, G., Carballo, S., Bergenstahl, B., & Tornberg, E. (2013). Dietary fiber, fructooligosaccharides, and physicochemical properties of homogenized aqueous suspensions of yacon (*Smallanthus sonchifolius*). *Food Research International*, 50(1), 392–400.
- Charles, A. L., Sriroth, K., & Huang, T. -c. (2005). Proximate composition, mineral contents, hydrogen cyanide and phytic acid of 5 cassava genotypes. *Food Chemistry*, 92(4), 615–620.
- Choque Delgado, G., da Silva Cunha Tamashiro, W., Maróstica Junior, M., & Pastore, G. (2013). Yacon (*Smallanthus sonchifolius*): A functional food. *Plant Foods for Human Nutrition*, 1–7.
- Colombo, A., Pérez, G. T., Ribotta, P. D., & León, A. E. (2008). A comparative study of physicochemical tests for quality prediction of Argentine wheat flours used as corrector flours and for cookie production. *Journal of Cereal Science*, 48(3), 775–780.
- Dell'Agli, M., Buscicala, A., & Bosio, E. (2004). Vascular effects of wine polyphenols. *Cardiovascular Research*, 63(4), 593–602.
- Dini, C., García, M. A., & Viña, S. Z. (2012). Non-traditional flours: frontiers between ancestral heritage and innovation. *Food & Function*, 3(6), 606–620.
- Doporto, M. C., Dini, C., Mugridge, A., Viña, S. Z., & García, M. A. (2012). Physicochemical, thermal and sorption properties of nutritionally differentiated flours and starches. *Journal of Food Engineering*, 113(4), 569–576.
- Doporto, M. C., Mugridge, A., García, M. A., & Viña, S. Z. (2011). *Pachyrhizus ahipa* (Wedd.) Parodi roots and flour: Biochemical and functional characteristics. *Food Chemistry*, 126(4), 1670–1678.

- Dragsted, L. O., Strube, M., & Larsen, J. C. (1993). Cancer-protective factors in fruits and vegetables: Biochemical and biological background. *Pharmacology & Toxicology*, 72(Suppl. 1), 116–135.
- Dreher, M. L. (2001). Chapter 1 dietary fiber overview. In S. Sungsoo Cho, & M. L. Dreher (Eds.), *Handbook of dietary fiber*. New York: Marcel Dekker, Inc.
- Duyvejonck, A. E., Lagrain, B., Dornez, E., Delcour, J. A., & Courtin, C. M. (2012). Suitability of solvent retention capacity tests to assess the cookie and bread making quality of European wheat flours. *LWT – Food Science and Technology*, 47(1), 56–63.
- Duyvejonck, A. E., Lagrain, B., Pareyt, B., Courtin, C. M., & Delcour, J. A. (2011). Relative contribution of wheat flour constituents to Solvent Retention Capacity profiles of European wheats. *Journal of Cereal Science*, 53(3), 312–318.
- Eluagu, E. N., & Onimawo, I. A. (2010). Effect of processing on the mineral composition and antinutritional factors of orange fleshed sweet potato (*Ipomoea batatas* L. Lam) flours. *Electronic Journal of Environmental, Agricultural and Food Chemistry*, 9(6), 1000–1005.
- Gaines, C. S. (2000). Report of the AACC committee on soft wheat flour. Method 56-11, Solvent Retention Capacity Profile. *Cereal Foods World*, 45, 303–306.
- García-Estépa, R. M., Guerra-Hernández, E., & García-Villanova, B. (1999). Phytic acid content in milled cereal products and breads. *Food Research International*, 32(3), 217–221.
- Greseli, P., Cerletti, C., Guglielmini, G., Pignatelli, P., de Gaetano, G., & Violi, F. (2011). Effects of resveratrol and other wine polyphenols on vascular function: An update. *The Journal of Nutritional Biochemistry*, 22(3), 201–211.
- Gurzau, E. S., Neagu, C., & Gurzau, A. E. (2003). Essential metals – Case study on iron. *Ecotoxicology and Environmental Safety*, 56(1), 190–200.
- Guzmán-Maldonado, H., Castellanos, J., & De Mejía, E. G. (1996). Relationship between theoretical and experimentally detected tannin content of common beans (*Phaseolus vulgaris* L.). *Food Chemistry*, 55(4), 333–335.
- Hang, D. T., Vanhanen, L., & Savage, G. (2013). Effect of simple processing methods on oxalate content of taro petioles and leaves grown in central Viet Nam. *LWT – Food Science and Technology*, 50(1), 259–263.
- Harland, B. F., & Morris, E. R. (1995). Phytate: A good or a bad food component? *Nutrition Research*, 15(5), 733–754.
- Havas, S., Roccella, E. J., & Lenfant, C. (2004). Reducing the public health burden from elevated blood pressure levels in the United States by lowering intake of dietary sodium. *American Journal of Public Health*, 94(1), 19–22.
- Hidvegi, M., & Lasztity, R. (2002). Phytic acid content of cereals and legumes and interaction with proteins. *Periodica Polytechnica Series in Chemical Engineering*, 46, 59–64.
- Huang, C. -C., Chen, W. -C., & Wang, C. -C. R. (2007). Comparison of Taiwan paddy- and upland-cultivated taro (*Colocasia esculenta* L.) cultivars for nutritive values. *Food Chemistry*, 102(1), 250–256.
- Kakade, M. L., Simons, N., & Liener, I. E. (1969). An evaluation of natural vs synthetic substrates for measuring the antitryptic activity of soybean samples. *Cereal Chemistry*, 46, 518–526.
- Kikunaga, S., Takahashi, M., & Huzisige, H. (1985). Accurate and simple measurement of phytic acid contents in cereal grains. *Plant & Cell Physiology*, 26(7), 1323–1330.
- Leidi, E. O., Sarmiento, R., & Rodríguez Navarro, D. N. (2003). Ahipa (*Pachyrhizus ahipa* [Wedd.] Parodi): An alternative legume crop for sustainable production of starch, oil and protein. *Industrial Crops and Products*, 17(1), 27–37.
- Leonel, M., Sarmiento, S. B. S., Cereda, M. P., & Câmara, F. L. A. (2003). Extração e caracterização de amido de jacatupé (*Pachyrhizus ahipa*). *Ciência e Tecnologia de Alimentos*, 23, 362–365.
- Leterme, P., Buldgen, A., Estrada, F., & Londoño, A. M. (2006). Mineral content of tropical fruits and unconventional foods of the Andes and the rain forest of Colombia. *Food Chemistry*, 95(4), 644–652.
- Lewu, M. N., Adebola, P. O., & Afolayan, A. J. (2010). Effect of cooking on the mineral contents and anti-nutritional factors in seven accessions of *Colocasia esculenta* (L.) Schott growing in South Africa. *Journal of Food Composition and Analysis*, 23(5), 389–393.
- López, O. V., Viña, S. Z., Pachas, A. N. A., Sisterna, M. N., Rohatsch, P. H., Mugridge, A., et al. (2010). Composition and food properties of *Pachyrhizus ahipa* roots and starch. *International Journal of Food Science and Technology*, 45(2), 223–233.
- Makkar, H. P. S., Blümmel, M., Borowy, N. K., & Becker, K. (1993). Gravimetric determination of tannins and their correlations with chemical and protein precipitation methods. *Journal of the Science of Food and Agriculture*, 61(2), 161–165.
- Makkar, H. P., Siddhuraju, P., & Becker, K. (2007). Plant secondary metabolites. *Methods in Molecular Biology*, 393, 1–122.
- N.R.C. (1989). *Lost crops of the Incas: Little-known plants of the Andes with promise for worldwide cultivation*. Washington DC: National Academy Press.
- Novak, W. K., & Haslberger, A. G. (2000). Substantial equivalence of antinutrients and inherent plant toxins in genetically modified novel foods. *Food and Chemical Toxicology*, 38(6), 473–483.
- Oboh, G., & Akindahunsu, A. A. (2003). Biochemical changes in cassava products (flour & gari) subjected to *Saccharomyces cerevisiae* solid media fermentation. *Food Chemistry*, 82(4), 599–602.
- Opara, L. (2003). Postharvest technology of root and tuber crops. In R. Dris, R. Niskanen, & S. Mohan Jain (Eds.), *Crop management and postharvest handling of horticultural products, Vol. II*. (pp. 381–406) Enfield, NH, USA: Science Publishers Inc.
- Ørting, B., Grüneberg, W. J., & Sørensen, M. (1996). Ahipa (*Pachyrhizus ahipa* (Wedd.) Parodi) in Bolivia. *Genetic Resources and Crop Evolution*, 43(5), 435–446.
- Phillippy, B. Q., Lin, M., & Rasco, B. (2004). Analysis of phytate in raw and cooked potatoes. *Journal of Food Composition and Analysis*, 17(2), 217–226.
- Ramos-de-la-Peña, A. M., Renard, C. M. G. C., Wicker, L., & Contreras-Esquivel, J. C. (2013). Advances and perspectives of *Pachyrhizus* spp. in food science and biotechnology. *Trends in Food Science & Technology*, 29(1), 44–54.
- Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology & Medicine*, 26(9–10), 1231–1237.
- Rimbach, G., & Pallauf, J. (1998). Phytic acid inhibits free radical formation in vitro but does not affect liver oxidant or antioxidant status in growing rats. *The Journal of Nutrition*, 128(11), 1950–1955.
- Rondán-Sanabria, G. G., Valcarcel-Yamani, B., & Finardi-Filho, F. (2012). Effects on starch and amylolytic enzymes during *Lepidium meyenii* Walpers root storage. *Food Chemistry*, 134(3), 1461–1467.
- Rosell, C. M., Santos, E., & Collar, C. (2009). Physico-chemical properties of commercial fibres from different sources: A comparative approach. *Food Research International*, 42(1), 176–184.
- Saris, N. E., Mervaala, E., Karppanen, H., Khawaja, J. A., & Lewenstam, A. (2000). Magnesium. An update on physiological, clinical and analytical aspects. *Clinica Chimica Acta*, 294(1–2), 1–26.
- Sasaki, M., Murakami, M., Matsuo, K., Matsuo, Y., Tanaka, S., Ono, T., et al. (2008). Oxalate nephropathy with a granulomatous lesion due to excessive intake of peanuts. *Clinical and Experimental Nephrology*, 12(4), 305–308.
- Schelp, F. -P., & Pongpaew, P. (1988). Protection against cancer through nutritionally-induced increase of endogenous proteinase inhibitors – A hypothesis. *International Journal of Epidemiology*, 17(2), 287–292.
- Shewry, P. R. (2003). Tuber storage proteins. *Annals of Botany*, 91(7), 755–769.
- Smith, C., Van Megen, W., Twaalfhoven, L., & Hitchcock, C. (1980). The determination of trypsin inhibitor levels in foodstuffs. *Journal of the Science of Food and Agriculture*, 31(4), 341–350.
- Sreeramulu, D., & Raghunath, M. (2010). Antioxidant activity and phenolic content of roots, tubers and vegetables commonly consumed in India. *Food Research International*, 43(4), 1017–1020.
- Steinmetz, K. A., & Potter, J. D. (1996). Vegetables, fruit, and cancer prevention: A review. *Journal of the American Dietetic Association*, 96(10), 1027–1039.
- Vaintraub, I. A., & Lapteva, N. A. (1988). Colorimetric determination of phytate in unpurified extracts of seeds and the products of their processing. *Analytical Biochemistry*, 175(1), 227–230.
- Wu, P., Tian, J. -C., Walker, C. E., & Wang, F. -C. (2009). Determination of phytic acid in cereals – A brief review. *International Journal of Food Science and Technology*, 44(9), 1671–1676.