LWT - Food Science and Technology 45 (2012) 94–102

Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt

Physicochemical and functional characterization of by-products from chia (*Salvia hispanica* L.) seeds of Argentina

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ARTICLE INFO

Article history:
Received 30 September 2010
Received in revised form
6 July 2011
Accepted 8 July 2011

Keywords:
Salvia hispanica
Chia
By-product
Physicochemical
Functional properties

ABSTRACT

The objective of this study was to characterize the physicochemical and functional properties of meals (M) and fibrous fractions (FRF) of chia seeds (*Salvia hispanica* L.), and to compare the effect of oil extraction methods (pressing -p- and solvent extraction -s-) and sieving process on these properties. Both processes affect the physicochemical and functional properties of residual meals and their corresponding fibrous fractions. Mp and FRFp showed a significantly higher residual oil content than Ms and FRFs (11.39, 10.85, 0.21 and 0.21 g/100 dry base, respectively). The sieving process of both meals allowed to obtain fibrous fractions with a significant increase of crude fiber (27.57, 32.84, 23.81 and 28.35 g/100 g in Ms, FRFs, Mp and FRFp, respectively), and a marked decrease of protein content (41.36, 35.32, 35.00 and 33.74 g/100 g in Ms, FRFs, Mp and FRFp, respectively). Total dietary fiber and their respective components (soluble and insoluble dietary fiber) were significantly higher in FRF. All the samples exhibited a high antioxidant activity due to the presence of phenolic compounds and tocopherols in the case of Mp and FRFp. Ms and FRFs presented a better oil-holding capacity, organic molecule absorption capacity, emulsifying activity and emulsion stability than Mp and FRFp, and allowed to achieve more stable emulsions. FRFs showed the highest values of water absorption and adsorption capacity.

1. Introduction

Chia (*Salvia hispanica* L.) is an annual herbaceous plant that belongs to the *Lamiaceae* family, which is native from southern Mexico and northern Guatemala. Chia seed together with corn, beans, and amaranth, were important crops for pre-Columbian civilizations in America, including the Mayan and Aztec populations (Álvarez-Chávez, Valdivia-López, Alberto-Juárez, & Tecante, 2008). Chia is considered an alternative crop to diversify and stabilize the economy of north-western Argentina (Coates & Ayerza, 1996). The plant produces numerous small white and dark seeds that mature in autumn. The seed has about 30 g oil/100 g seed weight, and it is mainly composed of unsaturated fatty acids (Bushway, Belyea, & Bushway, 1981; Taga, Miller, & Pratt, 1984). Chia seeds from Argentina exhibited 30.0–38.6 g oil/100 g, with 60.7–67.8 g/100 g of linolenic acid (Coates & Ayerza, 1996; Ixtaina et al., 2011).

The chia seed is a good source of protein (19–27 g/100 g) (Weber, Gentry, Kohlhepp, & McCrohan, 1991). The protein content is higher than that of other traditional crops such as wheat, corn, rice, oat, barley and amararanth (Ayerza & Coates, 2005). Although chia is not commercially grown as a protein source, its amino acid profile has no limiting factors in the adult diet (Bushway et al., 1981), but threonine, lysine and leucine were the limiting amino acids in a preschool child's diet (Weber et al., 1991).

The residual meal from the oil-extracting process of chia seeds is a good source of dietary fiber (Reyes-Caudillo, Tecante, & Valdivia-López, 2008) and polyphenolic compounds with antioxidant activity (Taga et al., 1984). This seed could be used as a source of important natural antioxidants with commercial applications (Reyes-Caudillo et al., 2008).

The consumption of chia dietary fiber can be an important alternative to improve people's health. Vázquez-Ovando, Rosado-Rubio, Chel-Guerrero, and Betancur-Ancona (2009) evaluated the physicochemical and physiological properties of a fibrous fraction obtained from Mexican chia seeds defatted by solvent extraction, suggesting its use as an ingredient in health and diet food products.

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doi:10.1016/j.lwt.2011.07.012

Dietary fiber (DF) includes cellulose, hemicellulose, lignin, pectins, gums, mucilage and other polysaccharides and oligosaccharides associated with plants. It is resistant to digestion and absorption in the human small intestine, with complete or partial fermentation in the large intestine (Esposito et al., 2005). Fibers have been classified as soluble (SDF), such as viscous or fermentable fibers which are fermented in the colon, and insoluble fibers (IDF), that have bulking action with limited extent fermentation in the colon (Anderson et al., 2009).

The addition of dietary fiber can affect food texture, playing a role as texturing and stabilizing agent. SDF contributes to the stabilization of food structure (dispersions, emulsions, etc.) through gel formation or thickening of the continuous phase. On the other hand, IDF increases the firmness of the products and provides a high fat absorption capacity (Oreopoulou & Tzia, 2007).

The knowledge about functional properties such as water-holding, absorption and adsorption capacity, as well as those linked to the affinity for lipid components, is useful for the food industry, from manufacture to final destination of the product and marketing conditions (Zambrano, Meléndez, & Gallardo, 2001). In this way, fibers are added to cooked meat products in order to increase the cooking yield due to their water and fat retention properties. On the other hand, in fried food products, the addition of fiber reduces lipid retention and increases moisture content (Raghavendra et al., 2006).

The properties of DF may be affected by its chemical form and by the conditions of the processes for obtaining food. Thus, the milling process can be modulated to obtain dietary fiber-rich cereal by-products and also to increase the SDF/IDF ratio. On the other hand, heat treatments have a strong impact on the chemical composition and physical properties of cereal dietary fiber (Vitaglione, Napolitano, & Fogliano, 2008). Chang and Morris (1990) found that heat treatments could affect the properties of DF in various foods such as apples, oat bran and corn, among others. Gualberto, Bergman, Kazemzadeh, and Weber (1997) investigated the effect of extrusion on the DF and phytic acid in cereal brans; they found a decrease in the IDF content during extrusion cooking, and an increase in the amount of SDF. It has also been reported that the hydration properties of DF might be modified by grinding, increasing the ability to retain water within the matrix of the fiber (Raghavendra et al., 2006).

The preparation of the raw materials and the different processes of oil extraction from the seeds (pressing and solvent extraction) could affect differently the cell structure and the residual oil content of the meal. In addition, the milling process can influence the physicochemical, functional and physiological properties. Ixtaina et al. (2011) observed that both extraction methods affected significantly the oil yield, and the quality and composition of some minor constituents of chia seed oil.

The fractionation of meals by dry sieving is a method that allows to obtain fractions with different chemical compositions, and by-products rich in fiber, protein and starch (Otto, Baik, & Czuchajowska, 1997; Vázquez-Ovando et al., 2009; and Vázquez-Ovando, Rosado-Rubio, Chel-Guerrero, & Betancur-Ancona, 2010).

The purpose of this study was to characterize the physicochemical and functional properties of meals and fibrous fractions of chia seeds, and to compare the effect of oil extraction methods (pressing and solvent extraction) and sieving processes on these properties.

2. Materials and methods

2.1. Seeds

Commercial chia seeds used in this study were obtained from Salta (25° south and 65.5° west), Argentina (10 kg). Seeds were packed in hermetic plastic vessels and stored at 5 °C until use.

2.2. Meals

Meals were obtained as by-products of two oil extraction methods:

2.2.1. Solvent extraction

Meal (Ms) was obtained after oil solvent extraction (*n*-hexane) in a Soxhlet apparatus (Buenos Aires, Argentina) by thermal cycles at 80 °C for 8 h, following the IUPAC Standard Method (1992), of chia seed previously ground in a laboratory mill (Moulinex, horizontal blade grinder, Buenos Aires Argentina) at 424 μm.

2.2.2. Pressing extraction

Seeds were pressed at 25–30 °C using a Komet screw press (Model CA 59 G, IBG Monforts, Mönchengladbach, Germany), with a 5-mm restriction dye and a screw speed of 20 rpm. The screw press was first run for 15 min without seed material, but with heating *via* an electrical resistance-heating ring attached around the press barrel to raise the screw press barrel temperature to the desired temperature (25 ± 2 °C). Running temperature was checked with a digital thermometer inserted in the restriction dye. The meal obtained by pressing (Mp) was ground with a laboratory mill (Moulinex, horizontal blade grinder, Buenos Aires, Argentina) at 498 μm.

Meals from both extraction methods (Ms and Mp) were homogenized and stored in plastic vessels at 5 °C until use.

2.3. Fibrous fractions

The fibrous fractions were obtained by dry fractionation of defatted meals (Otto et al., 1997; Vázquez-Ovando et al., 2009; and Vázquez-Ovando et al., 2010). The meals (Ms and Mp) were sieved in a Zonitest agitator (Buenos Aires, Argentina), using a 100 ASTM mesh (149 μm) for 20 min. The material retained by the sieve was considered rich in fiber (FRFs and FRFp, respectively).

2.4. Characterization of meals and fibrous fractions

The characterization of chia meals and fibrous fractions was carried out through the following determinations:

2.4.1. Particle size distribution

All samples were submitted to granulometry determination in a Zonytest agitator (Buenos Aires, Argentina) (equipped with 10, 14, 20, 35, 60, 100, 140, 200 and 325 ASTM meshes) for 60 min. The material retained by each sieve was weighed and the percentage of each fraction calculated.

The following fraction representing a different size particle: A (<44 μm), B (44–74 μm), C (74–105 μm), D (105–149 μm), E (149–250 μm), F (250–500 μm), G (500–840 μm), H (840–1410 μm), I (1410–2000 μm) and J (>2000 μm).

2.4.2. Proximate composition

AOCS (1998) procedures were used to determine moisture (method Ba 2^a-38), crude fiber (method Ba 6-84) and ash (method Ba 5^a-49). Oil (IUPAC 1.122, 1992) and nitrogen content (N_2) (AOAC, 1997; Guiragossian, Van Scoyoc, & Auxtell, 1979). Protein content was calculated as nitrogen × 6.25. Carbohydrate content was estimated as nitrogen-free extract (NFE) and these were calculated by difference using Eq. (1).

$$\text{NFE} = 100 - (\text{oil} + \text{protein} + \text{crude fiber} + \text{ash}) \quad (1)$$

2.4.3. Total (TDF), soluble (SDF) and insoluble (IDF) dietary fiber

Total dietary fiber (TDF), soluble dietary fiber (SDF) and insoluble dietary fiber (IDF) were determined according the gravimetric enzymatic method (Prosky, Asp, Schweizer, Devries, & Furda, 1988).

2.4.4. Neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin, cellulose and hemicellulose

The vegetable cell was separated into two parts (Van Soest method): cell content (highly digestible) and cell wall (partially digestible). The cell wall was analyzed and its components (cellulose, hemicellulose and lignin) were determinated. The technique makes use of acidic and neutral detergent (AOAC, 1997, Method 973.18; Guiragossian et al., 1979).

2.4.5. Minerals

Calcium, magnesium, iron, copper and zinc were measured by flame atomic absorption spectrophotometry using a GBC 902 Atomic Absorption Spectrometer (GBC Scientific Equipment, Victoria, Australia). Samples were mineralized (550 °C, 2 h) to obtain carbon free white ashes. Ashes were dissolved in chlorhydric acid, filtered and analyzed. Results were expressed as mg/kg meals or fibrous fractions.

Phosphorus content was determined following method AOCS Ca 12–55 (1998). This method determines phosphorus or equivalent phosphatide content by ashing the sample in the presence of zinc oxide, followed by the spectrometry measurement of phosphorus as a blue phosphomolybdic acid complex.

2.4.6. Antioxidant components

2.4.6.1. Polyphenol analysis. Polyphenolic compounds were extracted by three extractions: firstly, 10 mL of acetonitrile: 10 mL/100 mL acetic acid (50:50); secondly, 10 mL of acetonitrile: 10 mL/100 mL acetic acid (50:50); and thirdly, 10 mL of acetonitrile: 10 mL/100 mL acetic acid (70:30) and stirring by vortex. Samples were centrifuged at 498 g for 10 min (Centrifuge Sorvall Instruments RC3C, NY, USA) and the pooled extracts were evaporated to dryness by using a speed-vac evaporator Heto VR-1. The residue was dissolved in acetonitrile: 10 mL/100 mL acetic acid (50:50) and analyzed by HPLC-MS.

HPLC-MS analysis was performed using a Hewlett Packard 1050 system (Avondale, PA, USA), quaternary pump, with Metasil 3 μm ODS 150 × 4.6 mm column from Varian, and guard column Widepore C_{18} ODS 150 × 4.6 mm from Phenomenex (CA, USA). The mobile phase was acetonitrile: 10 mL/100 mL acetic acid (50:50). Flow rate was set at 1 mL/min; temperature was mantained at 35 °C. Peak detection was made by checking MS absorption at 323 nm (0–5 min) and 375 nm (5–20 min).

Polyphenolic compounds were identified by comparing their retention times with those of authentic standards: 4.35 min for chlorogenic acid, 3.6 min for caffeic acid, 9.8 min for myricetin, 11.8 min for quercetin and 13.3 min for kaempferol, and quantified using external standard.

2.4.6.2. Tocopherol analysis. Tocopherol content was determined on the lipid fraction of chia meals (Ms, Mp) and fibrous fractions (FRFs, FRFp) obtained by solvent extraction (*n*-hexane, Soxhlet at 80 °C for 8 h) (IUPAC Standard Method, 1992).

Tocopherol content was determined by normal phase HPLC using a Hewlett Packard chromatography system (HPLC Hewlett Packard 1050 Series, Waldbronn, Germany) following the procedures described in IUPAC 2.432 (IUPAC, 1992) and AOCS Ce 8-89 (1998). Approximately 0.25 g oil in 5 mL hexane was placed in an ultrasonic bath for 2 min and protected from light. A 20 μL aliquot of this solution was injected into a LiChrosorb Si 60 column (5 μm, 25 cm × 4.00 mm, Merck, Darmstadt, Germany) using n-hexane: isopropanol (99.5: 0.5, HPLC solvent, J.T. Baker, Phillpsburg, USA) as mobile phase at a flow rate of 1.5 mL min^{-1}. Tocopherols were detected using a fluorescence detector (Agilent, 1100 Series Fluorescence Detector G1321A, Palo Alto, CA, USA) with the excitation/emission wavelength set at 290/330 nm, and quantified using a six-point external standard curve.

2.4.7. Antioxidant activity

Extraction of phenolic compounds was carried out according to the method of Re et al. (1999). Ten mL ethanol were added to 1 g sample, then it was homogenized in Vortex for 2 min, decanted and filtered (0.45 μm nylon paper). The supernatant was transferred into a flask and evaporated using a rotavapor apparatus (BUCHI R-124, Germany) to concentrate the sample. It was then redissolved in 1000 μL ethanol.

A spectrophotometric method was employed to determine the antioxidant activity using a spectrophotometer (Hitachi U-1900 UV–VIS, Japan). Antioxidant activity was quantified by a dying assay of the radical cation $ABTS^{+}$, measuring $ABTS^{+}$ reduction as the percentage of absorption inhibition at 734 nm, just 6 min later. The radical cation ABTS and potassium persulfate were obtained from Sigma Aldrich. Chlorogenic acid was used as standard antioxidant. Results were expressed as μmol/L Trolox g/sample, considering that chlorogenic acid diminishes twice the amount of absortion than Trolox (Walker & Everette, 2009).

2.4.8. Functional properties

2.4.8.1. Water-holding (WHC) and oil-holding capacity (OHC). Both capacities were determined according to the method of Chau, Cheung, and Wong (1997). Briefly, 1 g (dry base (d.b.)) sample was weighed and then stirred into 10 mL distilled water or corn oil (density 0.92 g/mL, Arcor). These suspensions were centrifuged at 2200 × g for 30 min (Rolco Centrifuge Refrigerate, Model CR-5850, 22-cm radius, Buenos Aires, Argentina) and the supernatant volumes were measured. Water-holding capacity was expressed as g water held per g sample, and oil-holding capacity as g oil held per g sample.

2.4.8.2. Water adsorption capacity (WA_dC). This property was determined according to the method of Chen, Piva, and Labuza (1984). Briefly, 1 g (d.b.) sample was placed in an equilibrium micro-environment at 98% relative humidity, generated by placing 20 mL saturated potassium sulfate saline solution and placing these in oven at 25 °C until constant weight. Water adsorption capacity was expressed as g water per g sample.

2.4.8.3. Water absorption capacity (WA_bC). This property was determined according to the AACC (1984) method 88-04. Approximate water absorption capacity was first determined by weighing out 2 g (d.b.) sample, adding water until saturation (approx. 35 mL) and centrifuging at 2000 × g for 10 min in a Rolco Model CR-5850, radius 22 cm centrifuge (Buenos Aires, Argentina). Approximate water absorption capacity was calculated by dividing the increase in sample weight (g) quantifing the water needed to complete the original sample weight (2 g d b.) to 15 g. Water absorption capacity (WA_bC) was then determined by placing samples in four tubes, adding different quantities of water to bracket the measurement (1.5 and 0.5 mL water above original weight, and 1.5 and 0.5 mL water below; one in each tube), agitating vigorously, and centrifuging at 2000 × g for 10 min in a Rolco Model CR-5850, radius 22 cm centrifuge (Buenos Aires, Argentina). The supernatant was discarded and the residue weighed. Average water absorbed was calculated and the WA_bC calculated, and expressed as g water absorbed per g sample.

2.4.8.4. Organic molecule absorption capacity (OMAC). This capacity was determined according to the method of Zambrano et al. (2001). Three g (d b.) sample was placed in excess quantity corn oil (approx. 25 mL) for 24 h at room temperature, and then centrifuged at 2000 × g for 15 min in a Rolco Model CR-5850, radius 22 cm centrifuge (Buenos Aires, Argentina). Organic molecule absorption capacity was expressed as the absorbed hydrophobic component and calculated in terms of sample weight gain (g oil/sample g).

2.4.8.5. Emulsifying activity (EA) and emulsion stability (ES). These properties were evaluated according to the method of Chau et al. (1997). Briefly, 100 mL 2 g/100 mL suspension were homogenized using Ultra-Turrax T25 (Janke & Kunkel, IKA- Labortechnik, Germany) at 7800 rpm for 2 min. Then, 100 mL corn oil (density 0.92 g/mL, Arcor) were added and homogenized at 15,000 rpm for 2 min. Emulsions were centrifuged in a 15 mL graduated centrifuge tube at 455 × g for 10 min, and emulsion volume was measured. Emulsifying activity was expressed as the mL of the emulsified layer volume of the 100 mL entire layer in the centrifuge tube. Emulsion stability was determined by heating the emulsions to 80 °C for 30 min, cooling them to room temperature and centrifuged at 455 g for 10 min. Emulsion stability was expressed as mL of the remaining emulsified layer volume of 100 mL the original emulsion volume.

On the other hand, all emulsions were evaluated by optical characterization using a Vertical Scan Analyzer (QuickSCAN, Beckman Coulter, Fullerton, USA). The QuickSCAN head scans the entire length of the sample (about 65 mm), acquiring backscattering (BS) data every 40 μm. Thus, it is possible to obtain curves giving the backscattering light flux as percentage, relative to external standards, as a function of the sample height in mm (Pan, Tomás, & Añón, 2002). Coalescence kinetics were followed by measuring the mean values of BS as a function of time in the zone of 25–30 mm (Backscattering % 25–30 mm).

2.5. Statistical analysis

All determinations were done in triplicate. Results were analyzed using ANOVA test followed by Tukey's test ($p < 0.05$), using Infostat software (Infostat Group, Facultad de Ciencias Agrarias, Universidad Nacional de Córdoba, Argentina, 2004).

3. Results and discussion

The seed oil extraction process produced 80.8 ± 0.3 (g/100g d.b.) residual meal by solvent (Ms) and 75.2 ± 0.5 (g/100g d.b.) residual meal by pressing (Mp), whereas the sieving procedure yielded 79.9 ± 0.8 g/100 g and 85.8 ± 0.5g/100 g fibrous fractions (FRFs and FRFp, respectively).

3.1. Granulometric analysis

Fig. 1 shows the analysis of chia meal particle size corresponding to solvent and pressing extraction processes (Ms and Mp), as well as their fibrous fractions (FRFs and FRFp), respectively. Ms showed a higher percentage of smaller particle size (20.1g/100 g) (ranging from <44 to 149 μm) than that shown by Mp (14.1 g/100 g), whereas fibrous fractions in both residual meals only presented traces of those particles which could be distributed in Ms through sieving. However, in Mp they could only be retained in the larger size fraction because the presence of residual lipids could have affected particle agglomeration.

Samples corresponding to the solvent extraction process presented the largest particle size from 149 to 840 μm (79.5 and 98.5 g/100 g in Ms and FRFs, respectively), whereas those corresponding to the pressing extraction were between 250 and 840 μm (82.3 and 95.3 g/100 g in Mp and FRFp, respectively). The comparison of fibrous fractions showed that the FRFp had a higher proportion of larger particles (>250 μm) (85.8 and 98.4 g/100 g in FRFs and FRFp, respectively).

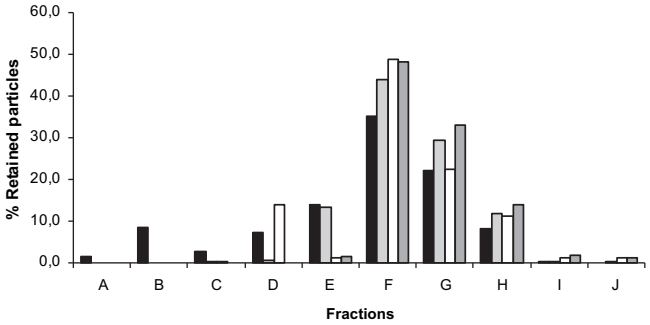

Fig. 1. Particle size distribution of chia (*Salvia hispanica* L.) meals and fibrous fractions. Each value is an average of three determinations ($n = 3$). A: <44 μm; B: 44–74 μm; C: 74–105 μm; D: 105–149 μm; E: 149–250 μm; F: 250–500 μm; G: 500–840 μm; H: 840–1410 μm; I: 1410–2000 μm and J: > 2000 μm ■ = Ms; ▨ = FRFs; □ = Mp; ▩ = FRFp.

3.2. Proximate composition

Table 1 presents the percentage of each component analyzed in chia meals and fibrous fractions obtained by both extraction methods. Samples showed a high percentage of proteins and crude fiber. It must be noted that crude fiber content was determined by a proximate method that quantifies mainly cellulose. Ms and Mp exhibited a significantly higher protein content than FRFs and FRFp, respectively. Both meals (Ms and Mp) can be incorporated in human diets and mixed with other grains to produce a more balanced protein source (Ayerza & Coates, 2011). In turn, all the samples had a higher crude fiber content than that reported for canola, soybean and flax meals (11.54, 3.50 and 5.27 g/100 g, respectively). On the other hand, the meal obtained by solvent extraction was found to have a higher protein content than that corresponding to canola and flax meals (36.13 and 38.96 g/100 g, respectively; Khattab & Arntfield, 2009). Samples obtained by the pressing process (Mp and FRFp) showed a significantly higher residual oil content ($p < 0.05$) than that corresponding to solvent extracted samples.

On the other hand, the sieving process of both meals allowed obtaining fibrous fractions with a significant increase ($p < 0.05$) of crude fiber and a marked decrease ($p < 0.05$) of protein content. Thus, structural carbohydrates that are located on the vegetable

Table 1
Proximate composition of chia (*Salvia hispanica* L.) meals and fibrous fractions from different extraction methods (g/100 g d b).

Component	Ms	FRFs	Mp	FRFp
Moisture	10.47 ± 0.16	10.34 ± 0.03	10.84 ± 0.21	10.00 ± 0.16
Protein*	41.36 ± 0.28^{c}	35.32 ± 0.17^{b}	35.00 ± 0.35^{b}	3374 ± 0.05^{a}
Crude fiber	27.57 ± 0.07^{b}	32.84 ± 0.34^{c}	23.81 ± 0.34^{a}	28.35 ± 0.80^{b}
Oil	0.21 ± 0.08^{a}	0.21 ± 0.05^{a}	11.39 ± 0.59^{b}	10.85 ± 0.13^{b}
Ash	7.24 ± 0.15^{c}	6.64 ± 0.03^{b}	6.27 ± 0.08^{a}	6.04 ± 0.01^{a}
NFE	23.62 ± 0.94	24.99 ± 0.56	23.53 ± 0.87	21.02 ± 0.79

Values followed by different letters differ at $p < 0.05$, according to Tukey test.
Results are expressed as ($x \pm d$), being x the average and d, the standard deviation ($n = 3$).
Ms: solvent meal; FRFs: solvent fiber-rich fraction; Mp: pressing meal; FRFp: pressing fiber-rich fraction.
*Factor: 6.25; NFE: nitrogen-free extract.

cellular wall could be retained, and cellular components could be transferred (Pérez, 2001).

3.3. Total (TDF), soluble (SDF) and insoluble (IDF) dietary fiber

Meals (Ms and Mp) and fibrous fractions (FRFs and FRFp) of chia presented a high content of TDF, consisting mostly of IDF. TDF concentration and its corresponding components (SDF and IDF) were significantly higher in chia fibrous fractions ($p < 0.05$) (Table 2). The intake of appropriate quantities of DF is related to the prevention of chronic diseases such as hypercholesterolemia, diabetes, colon cancer, and obesity, among others (Lecumberri et al., 2007).

The American Diabetic Association recommends a daily fiber intake of 25–30 g, being the relation between IDF and SDF an important piece of information for the nutritional and physiological effects in consumers (Borderías, Sánchez-Alonso, & Pérez-Mateos, 2005). For the set of studied samples, the IDF/SDF ratio varied between 9.5 and 11 (Ms and FRFp values, respectively), lower than the value detected by Vázquez-Ovando et al. (2009) for a Mexican chia fiber fraction (17.1), and than that observed by Grigelmo-Miguel and Martín-Belloso (1999) for wheat bran (14.2). These values turn the chia by-products analyzed into an ingredient of great interest to be incorporated in several foods, due to the capacity of SDF to retain water and increase satisfaction after eating, as well as to decrease the time of nutrient absorption (Scheneeman, 1987). In addition, these by-products would act technologically as important thickening agents, gelling and stabilizers of foams and emulsions.

The content of TDF and IDF (d.b.) was not affected by the method to obtain meals (Table 2), where significant differences can be observed in relation with SDF. Such differences could be attributed to the higher content of residual lipids in Mp and FRFp with respect to Ms and FRFs (Table 1). However, by comparing the content of TDF expressed in dry base and oil free, a significant higher concentration (49.77 g/100 g d b oil free) was found in Mp with respect to Ms (46.02 g/100 g d b oil free). This increase in fiber can be associated with a decrease in protein content by its dragging with oil during pressing (39.50 g/100 g d b oil free and 41.45 g/100 g d b oil free for Mp and Ms, respectively).

3.4. Neutral detergent fiber, acid detergent fiber, cellulose, lignin and hemicellulose

For all the samples, NDF composed of cellulose, hemicellulose and lignin (structural polysaccharides and lignin that contribute to the IDF fraction and make it to increase its proportion of TDF) changed within the 59.3 and 64.9 g/100 g range. Chia fibrous fractions had a higher content (although statistically not significant, Fig. 2) of NDF and ADF compared to that of the corresponding meals, confirming that the material obtained through sieving was richer in fiber (Table 2). Similar results were reported for fine and coarse wheat bran by Kirwan, Smith, McConnell, Mitchell, and Eastwood (1974).

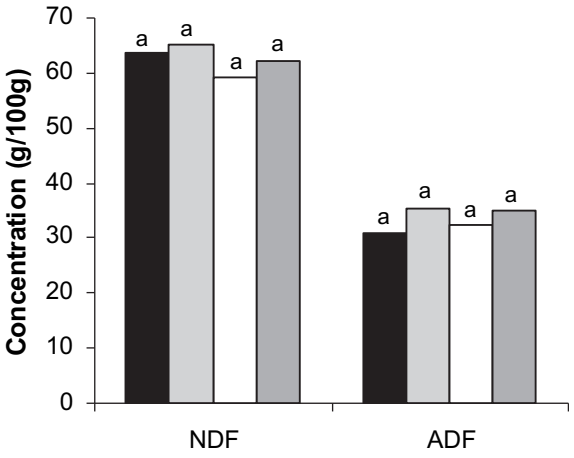

Fig. 2. Neutral and Acid detergent fiber (NDF) and acid detergent fiber (ADF) of meals and fibrous fraction of chia (*Salvia hispanica* L.). Each value is an average of three determinations ($n = 3$). ■ = Ms; ▒ = FRFs; □ = Mp; ▒ = FRFp.

Different components of NDF for the analyzed samples are shown in Table 3. It can be observed that both meals (Ms and Mp) did not present significant differences concerning cellulose, hemicellulose and lignin content. However, among the fibrous fractions there were no significant differences in lignin content, this component being statistically higher in FRFp. This fact could be related to the higher percentage of large particles in FRFp (98.4 and 85.8 g/100 g in FRFp and FRFs, respectively), since the presence of residual lipids could have affected particle agglomeration. Similar results were reported for coarse and fine wheat bran (4.1 and 2.6 g/100 g lignin, respectively) (Kirwan et al., 1974).

FRFs, on the other hand, exhibited a significantly higher content of cellulose than Ms, which could be attributed to the high amount of particles of smaller size in Ms (fine 20%).

3.5. Minerals

Phosphorus, calcium, magnesium, zinc, iron and copper were detected in all the samples, being phosphorus, calcium and magnesium in the highest proportion (Table 4). In turn, it can be observed that there was no clear trend concerning the mineral concentration in the analyzed samples. This concentration was higher than that reported for coarse wheat, rice and sorghum meals by Ragaee, Abdel-Aal, and Norman (2006), whereas iron content was higher than that of sesame, but similar to that of barley (Egbekun & Ehieze, 1997).

Solvent-extracted meal (Ms) was characterized by a high phosphorus and calcium concentration, in agreement with the

Table 2
Total (TDF), soluble (SDF) and insoluble (IDF) dietary fiber of chia (*Salvia hispanica* L.) meals and fibrous fractions from different extraction methods (g/100 g d b).

Component	Ms	FRFs	Mp	FRFp
TDF	45.92 ± 0.74^{a}	52.26 ± 0.65^{b}	44.11 ± 0.18^{a}	51.98 ± 0.20^{b}
IDF	41.55 ± 0.67^{a}	47.51 ± 0.59^{b}	40.30 ± 0.16^{a}	47.65 ± 0.18^{b}
SDF	4.37 ± 0.07^{b}	4.75 ± 0.06^{c}	3.81 ± 0.02^{a}	4.33 ± 0.02^{b}

Values followed by different letters differ at $p < 0.05$, according to Tukey test.
Results are expressed as ($x \pm d$), being x the average and d, the standard deviation ($n = 3$).
Ms: solvent meal; FRFs: solvent fiber-rich fraction; Mp: pressing meal; FRFp: pressing fiber-rich fraction.

Table 3.
Neutral detergent fiber composition (g/100 g b s) of chia (*Salvia hispanica* L.) meals and fibrous fractions of both extraction methods

Component	Ms	FRFs	Mp	FRFp
Celullose	23.0 ± 0.89^{a}	28.2 ± 0.43^{b}	22.0 ± 1.78^{a}	$25.4 \pm 1.44^{a,b}$
Hemicelullose	33.6 ± 0.99^{a}	31.3 ± 0.03^{a}	30.3 ± 1.99^{a}	29.9 ± 0.56^{a}
Lignin	$6.9 \pm 0.52^{a,b}$	5.5 ± 0.53^{a}	7.5 ± 0.08^{b}	8.1 ± 0.18^{b}

Values followed by different letters differ at $p < 0.05$, according to Tukey test.
Results are expressed as ($x \pm d$), being x the average and d, the standard deviation ($n = 3$).
Ms: solvent meal; FRFs: solvent fiber-rich fraction; Mp: pressing meal; FRFp: pressing fiber-rich fraction.

Table 4
Mineral content (mg/kg) of chia (*Salvia hispanica* L.) meals and fibrous fractions of both extraction methods (d b).

Element	Ms	FRFs	Mp	FRFp
Ca	8060 ± 0.05^{d}	6150 ± 0.05^{c}	5615 ± 0.05^{a}	6110 ± 0.05^{b}
Mg	3460 ± 0.03^{b}	3220 ± 0.03^{a}	4624 ± 0.02^{d}	3690 ± 0.02^{c}
Fe	117.3 ± 0.001^{a}	121.0 ± 0.001^{c}	117.7 ± 0.001^{b}	142.7 ± 0.001^{d}
Zn	100 ± 0.001^{d}	96 ± 0.001^{b}	6 ± 0.001^{c}	92.8 ± 0.001^{a}
Cu	24 ± 0.002^{c}	22.6 ± 0.002^{b}	18.7 ± 0.002^{a}	24.2 ± 0.02^{d}
P	10205 ± 1141^{a}	9012.5 ± 697^{a}	9988.5 ± 335^{a}	12476 ± 1505^{a}

Values followed by different letters differ at $p < 0.05$, according to Tukey test.
Results are expressed as $(x \pm d)$, being x the average and d, the standard deviation ($n = 3$).
Ms: solvent meal; FRFs: solvent fiber-rich fraction; Mp: pressing meal; FRFp: pressing fiber-rich fraction.

value reported by Bushway et al. (1981) for another chia meal (*Salvia polystachya*) which was obtained by a similar method.

The calcium/phosphorus ratio (Ca:P) in the samples analyzed was found to be within the range of 0.49–0.79; the lowest value corresponded to FRFp and the highest to Ms. That ratio was similar to that of a normal diet from the USA and Canada (Mota-Blancas & Perales-Caldera, 1999). Based on the facts that the Ca:P ratio must be within the range of 1:1 and 2:1 for an effective utilization by ruminants as well as to avoid nutritional disorders (FAO); that Ca absorption represents 20–30% of total ingestion, and that this process is affected by other food components such as proteins, fibers, phytic acid and phosphorus (Mota-Blancas & Perales-Caldera, 1999), chia by-products could be used as supplements in other cereal meals due to their high content of this mineral.Taking into account the high level of DF, a further analysis of dializability will allow to obtain further information in order to analyze the potential bioavailability of this mineral (Kernefick & Cashman, 2000).

3.6. Antioxidant components

3.6.1. Polyphenol analysis

Samples evidenced the presence of chlorogenic and caffeic acids, quercetin, myricetin and kaempferol with a total content of polyphenolic components ranging from 3.3×10^{-4} to 5.7×10^{-4} mol/kg, though no significant difference was detected in the total polyphenolic component concentration (Fig. 3). However, the samples corresponding to the solvent extraction process (Ms and FRFs) showed significantly higher concentrations ($p < 0.05$) of quercetin and myricetin than those obtained by pressing (Mp and FRFp). In turn, chlorogenic acid was found to be in higher concentrations in samples obtained by solvent extraction, even being statistically higher in FRFs than in those samples obtained by pressing ($p < 0.05$). On the other hand, pressing processed samples evidenced significantly higher concentrations of caffeic acid than those obtained by solvent extraction.

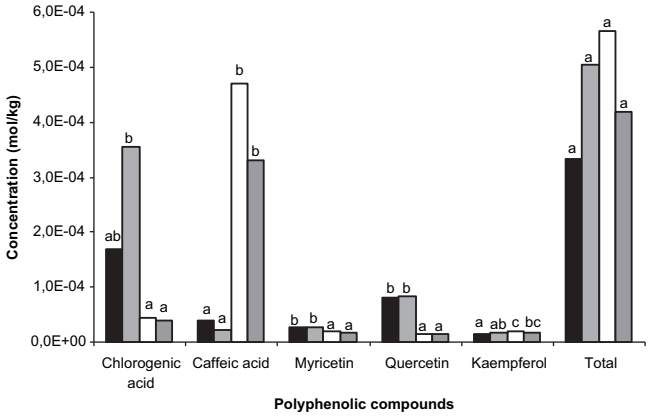

Fig. 3. Polyphenol components present in chia (*Salvia hispanica* L.) meals and fibrous fraction obtained by both extraction methods. Each value is an average of three determinations ($n = 3$). ■ = Ms; □ = FRFs; □ = Mp; ■ = FRFp.

3.6.2. Tocopherol analysis

Fig. 4 shows that meal and fibrous fraction obtained by pressing extraction (Mp and FRFp) had a total tocopherol concentration of approximately 470–545 mg/kg (Mp and FRFp, respectively), where γ-tocopherol was the main component (~95%), followed by α-and δ-tocopherol, but no β-tocopherol was detected. These samples showed a higher total tocopherol concentration than that of chia oil obtained by the same method (238 mg/kg) (Ixtaina et al., 2011). In fact, the pressing extraction process of oil was not proportional to that of tocopherols, which was found to be lower.

On the other hand, traces of tocopherols were found in the remaining solvent extracted samples (Ms and FRFs) (data not shown). This fact is related to the low lipid content of these samples (Table 1) and the high concentration of these components in chia oil obtained by solvent (300 mg/kg) (Ixtaina et al., 2011).

3.7. Antioxidant activity

Antioxidant activity of chia meals and fibrous fractions obtained by solvent and pressing extraction is shown in Table 5.

Samples corresponding to the pressing extraction method (Mp and FRFp) evidenced a greater antioxidant activity than that of samples obtained by solvent (Ms and FRFs). This may be related to the amount of tocopherols present in the samples as they are substances with natural antioxidant activity. On the other hand, in solvent extracted samples, the fibrous fraction showed a statistically higher antioxidant activity ($p < 0.05$) than the meal (Ms), whereas in those samples corresponding to pressing extraction method, the meal was found to have greater antioxidant activity. This fact could be due to a high content of polyphenolic components (Fig. 3).

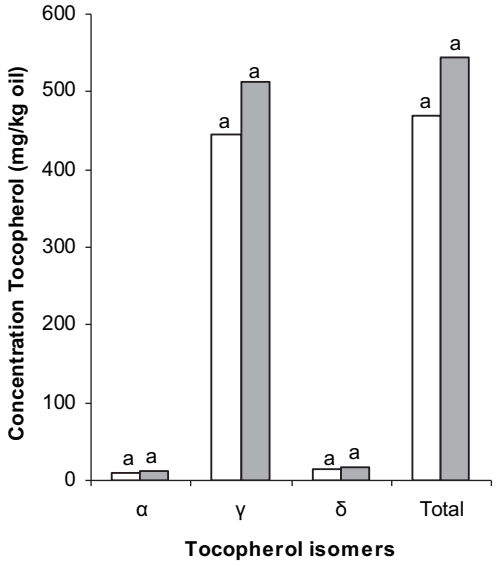

Fig. 4. Tocopherol content of lipid fraction of chia (*Salvia hispanica* L.) meal and fibrous fraction obtained by pressing extraction. Each value is an average of three determinations ($n = 3$). □ = Mp; ■ = FRFp.

Table 5
Antioxidant activity of chia (*Salvia hispanica* L.) meals and fibrous fractions of both extraction methods measured as $ABTS^{+}$ decolorization.

Sample	Trolox equivalent antioxidant coefficient (TEAC, μmol/g)
Ms	226.6 ± 4.13^{a}
FRFs	348.6 ± 51.77^{b}
Mp	557.2 ± 28.18^{c}
FRFp	446.4 ± 19.81^{b}

Values followed by different letters differ at $p < 0.05$, according to Tukey test.
Results are expressed as ($x \pm d$), being x the average and d, the standard deviation ($n = 3$).
Ms: solvent meal; FRFs: solvent fiber-rich fraction; Mp: pressing meal; FRFp: pressing fiber-rich fraction.

The antioxidant activity in all the samples was quite similar to that reported by Vázquez-Ovando et al. (2009) for a Mexican chia fibrous fraction (488.8 TEAC, μmol/g). The samples had a higher antioxidant activity than that found in wheat bran and sorghum and barley whole grain meals (48.5, 51.7 and 14.9 TEAC, μmol/g) (Iqbal, Bhanger, & Anwar, 2005; Ragaee et al., 2006).

3.8. Functional properties

Table 6 displays functional properties of chia meals and fibrous fractions obtained by both methods, solvent and pressing. Although the SDF content of all the chia seed by-products analized was comparatively low, they exhibited a high WHC. This may be due to the mucilages in the chia seeds that can act as SDF. Mucilages have excellent water-holding propierties. These mucilages do not quantify in the SDF because some components may not precipitate during the ethanol treatment for SDF determination and, therefore, SDF may be underestimated (Saura-Calixto & García-Alonso, 2001). Chia meals presented a similar capacity to retain, absorb and adsorb water (WHC, WA_bC and WA_dC, respectively). Their great capacity to retain and absorb water must be pointed out, being WA_bC greater than that of canola and soy meals (3.90 and 3.28 g/g, respectively), but similar to flax meal (6.03 g/g) (Khattab & Arntfield, 2009). On the other hand, both meals had a higher emulsifying activity than that of flax, soy and canola meals. Ms showed a similar OHC to that reported by Khattab and Arntfield (2009) for flax and canola meals (2.01 and 2.09 g/g, respectively), whereas Mp showed a lower capacity in those meals, and a significantly lower capacity compared to solvent extracted meal ($p < 0.05$) due to the high percentage of residual oil present in Mp (11.39 g/100 g) with respect to 0.21 g/100 g in Ms (Table 1). Likewise, both OMAC and the activity and emulsifying stability were significantly lower ($p < 0.05$) in the pressing extracted meal when compared to the solvent extracted one. This could be the result of a high percentage of residual lipids and a lower protein content in Mp.

Chia fibrous fractions showed functional features that were statistically different among them ($p < 0.05$). Nevertheless, both fibrous fractions evidenced a great capacity to retain and absorb water, and as an emulsifying and stabilizing agent of emulsions. Concerning the FRFs, functional properties, except water retention capacity, were significantly greater ($p < 0.05$) than those in FRFp. With respect to WHC, the differences found for the different fibrous fractions may be due to the influence of certain experimental parameters such as shaking, temperature, soaking time, nature and magnitude of the applies external force, particle size as well as environmentally chemical conditions (pH, ionic force). These factors could alter the fiber physical structure, leading to important changes in that property (Sangnark & Noomhorm, 2003). In both fractions, WHC was greater than the one corresponding to soy fiber (4.9 g/g), wheat bran (6.1 g/g) (Mongeau & Brassard, 1982) and corn and wheat husk (2.32 and 2–48 g/g, respectively) (Zambrano et al., 2001).

FRFs evidenced OHC similar to that of fibrous residues of barley (2.00 g oil/g sample) and of jack bean (2.3 g oil/g sample) (Betancur-Ancona, Peraza-Mercado, Moguer-Ordoñez, & Fuertes-Blanco, 2004), while WA_bC from that fraction was higher than the one reported by Zambrano et al. (2001) for carrot, prickly pear, cabbage, corn, wheat and soy husk (6.36; 5.75; 3.18; 3.17; 2.91 and 1.42 g/g, respectively), but similar to that of Mexican chia fibrous fraction (11.73 g/g) (Vázquez-Ovando et al., 2009). Water adsorption and organic molecule absorption in the FRFs were higher than the ones corresponding to the Mexican fibrous fraction (0.3 and 1.09 g/g WA_dC and OMAC, respectively) (Vázquez-Ovando et al., 2009). The WA_dC was similar to that of corn, soy and wheat husk (Zambrano et al., 2001). On the other hand, the emulsifying activity of FRFs was quite similar to that in the Mexican chia fibrous fraction. However, the emulsifying stability was lower than that of the Mexican species (94.84 mL/100 mL) (Vázquez-Ovando et al., 2009).

There were some differences between functional properties of meals and their fibrous fractions (Table 6), though sieving did not produce a clear trend in those properties with the exception of activity and stability of the emulsion. These properties were markedly higher than those in meals, which can be attributed to a greater proportion of proteins in the samples.

Destabilization kinetics of the corresponding emulsions in all analyzed samples is shown in Fig. 5. Initial stability (BSi) emulsions changed within the range of 60.7–67.2%, where the lowest value of BSi corresponded to FRFp emulsions and the highest one to FRFs. It must be noted that those emulsions from solvent extracted samples

Table 6
Functional properties of chia (*Salvia hispanica* L.) meals and fibrous fractions of both extraction methods.

Property	Ms	FRFs	Mp	FRFp
WHC (g/g)	10.64 ± 0.60^{b}	9.19 ± 0.29^{a}	10.58 ± 0.55^{b}	11.88 ± 0.33^{c}
OHC (g/g)	2.03 ± 0.08^{b}	2.06 ± 0.03^{b}	$1.26 \pm 0{,}03^{a}$	1.40 ± 0.18^{a}
WA_bC (g/g)	6.45 ± 0.41^{a}	10.46 ± 0.87^{b}	6.81 ± 0.30^{a}	6.13 ± 0.37^{a}
WA_dC (g/g)	0.37 ± 0.02^{a}	0.51 ± 0.05^{b}	0.31 ± 0.04^{a}	0.31 ± 0.03^{a}
OMAC (g/g)	1.64 ± 0.02^{b}	1.73 ± 0.05^{c}	0.83 ± 0.01^{a}	0.82 ± 0.01^{a}
EA (mL/100 mL)	56.00 ± 0.77^{d}	53.33 ± 0.00^{c}	51.00 ± 1.15^{b}	44.33 ± 1.15^{a}
ES (mL/100 mL)	60.00 ± 0.00^{d}	57.67 ± 1.15^{c}	47.17 ± 1.00^{b}	34.33 ± 1.15^{a}

Values followed by different letters differ at $p < 0.05$, according to Tukey test.
Results are expressed as ($x \pm d$), being x the average and d, the standard deviation ($n = 3$).
Ms: solvent meal; FRFs: solvent fiber-rich fraction; Mp: pressing meal; FRFp: pressing fiber-rich fraction.
WHC: water-holding capacity; OHC: oil-holding capacity; WAbC: water absorption capacity; WAdC: water adsorption capacity; OMAC: organic molecule absorption capacity; AA: antioxidant activity; EA: emulsifying activity and ES: emulsion stability.

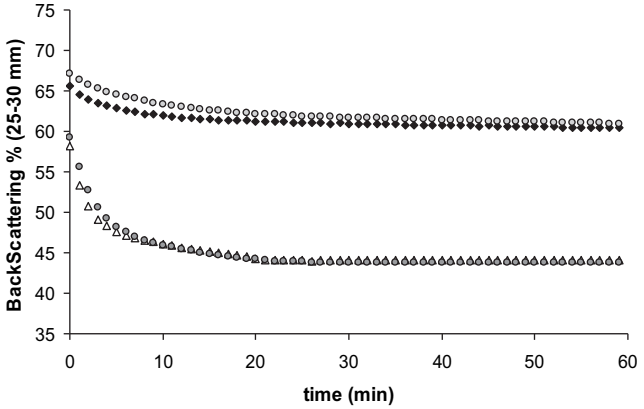

Fig. 5. Destabilization kinetics of emulsions prepared with meals and fibrous fraction of chia (*Salvia hispanica* L.) obtained by both extraction methods. Each value is an average of three determinations ($n = 3$). ◆ = Ms; ◯ = FRFs; △ = Mp; ● = FRFp.

(Ms and FRFs) were found to be quite stable; they slightly decreased around 5.5% BS along the period tested. This fact could be due to the high proportion of proteins and fibers present in these samples (Table 1), as they play the role of emulsifying and stabilizing agents. In contrast, emulsions corresponding to Mp and FRFp turned out to be less stable as 10 min later destabilization increased by 11.5%, reaching a total of 18%, due to a lower protein content and a higher residual percentage of oil.

4. Conclusions

Experimental results showed that the oil extraction methods (solvent and pressing) from chia seeds (*S. hispanica* L.) affect the physicochemical and functional properties of residual meals and their corresponding fibrous fractions. Meals and fibrous fractions of chia showed a high content of total dietary fiber (TDF), consisting mostly of insoluble dietary fiber (IDF). The concentration of TDF and its corresponding components (SDF and IDF) were significantly higher in the chia fiber fractions. All the samples exhibited a high antioxidant activity, which is associated with the polyphenolic compounds and the presence of tocopherols in the case of by-products Mp and FRFp.

The meal obtained by the solvent extraction method (Ms) evidenced significantly higher functional properties (OHC, OMAC, EA and ES) than the meal obtained by pressing extraction (Mp), whereas among the chia fibrous fractions (FRFs and FRFp), all the functional properties tested (WHC, OHC, WAbC, WAdC, OMAC, EA and ES) were statistically significant. Also, the emulsions formulated with by-products of chia obtained by solvent extraction were more stable than those obtained from pressing. These results could be attributed to the lower percentage of residual lipids and the higher protein content present in meals and fibrous fractions obtained by solvent extraction. They could also be explained in terms of the influence of certain experimental parameters that can alter the physical structure of the fiber, leading to important changes in these properties.

The physicochemical and functional properties turned chia by-products into important ingredients for the manufacture of products such as desserts, drinks, breads, jellies, emulsions, cookies, among others.

Acknowledgments

This work was supported by grants from PICT 2007-1085 (ANPCyT), CONICET PIP 1735-1085, Universidad Nacional de La Plata (UNLP, 11/X502) and Universidad Nacional del Centro de la Provincia de Buenos Aires (UNCPBA), Argentina. The authors thank Juan Facundo Massolo and Luís Rodoni for their technical assistance.

M.I. Capitani is a recipient of a fellowship from the Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET). Argentina. S.M. Nolasco is a Scientific and Technological Researcher and Professor at the Facultad de Ingeniería de la Universidad Nacional del Centro de la Provincia de Buenos Aires (UNCPBA). Argentina. M.C. Tomás is a member of the career of Scientific and Technological Researcher of the Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina.

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