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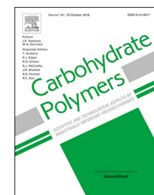
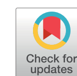


Effect of extraction temperature on rheological behavior and antioxidant capacity of flaxseed gum



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ABSTRACT

Soluble flaxseed gum (SFG) extracted at different temperatures (25, 40, and 60 °C) was analyzed in relation to the yield, polysaccharides and phenolics composition, surface charge, color, and rheological properties. The yield of SFG extract increased as the extraction temperature increased. The SFG xylan was the main component regardless the extraction temperature, but a reduction of substituents on the xylose chain was observed when increasing the extraction temperature. The phenolic compounds were also affected by the extraction temperature, influencing the antioxidant capacity of the gum. For all the extraction temperatures, SFG aqueous solutions showed a shear time-independent and shear-thinning behavior. Furthermore, oscillatory measurements showed a prevailing viscous character, but the decrease of the extraction temperature resulted in an increase of both G' and G''. Therefore, SFG extracted at low extraction temperatures showed higher viscous and elastic properties, while high extraction temperatures increased the antioxidant activity.

1. Introduction

The soluble portion of flaxseed (*Linum usitatissimum* L.) gum (SFG) or mucilage is contained mainly in the hull mucous epidermis. This polysaccharide can be easily extracted by soaking the flaxseed in water, and the efficiency of the process depends mainly on the temperature (Cui, Mazza, Oomah, & Biliaderis, 1994). SFG shows great potential to be used in the food industry owing to its sustainable, biodegradable and functional properties, and bio-safe characteristics. It can be used as a thickener or a stabilizer/emulsifier in food systems, promoting interesting texture/rheological properties due to its high water-solubility and structural interaction with other hydrocolloids such as starch, guar gum or proteins (Chen, Huang, Wang, Li, & Adhikari, 2016; Li, Li, Wang, Wu, & Adhikari, 2012; Wang et al., 2008), but its natural bioactive compounds can be equally useful for the enrichment of food products (Cui & Mazza, 1996; Cui, Mazza, & Biliaderis, 1994; Kennedy & Huang, 2003). The intake of SFG as dietary fiber can result in an improvement of the intestinal tract transit, reduced risk of diabetes and coronary heart diseases, decrease in the cholesterol and sugar absorption into the blood, decrease in the incidence of obesity, prevention of colorectal cancer, and other health benefits, such as help in treating the symptoms of depression, irritable bowel syndrome and osteoporosis (Liu, Shim, Poth, & Reaney, 2016; Mirhosseini & Amid, 2012; Morris & Vaisey-Genser, 2003).

The SFG is composed by a neutral and an acidic fraction of polysaccharides and proteins (Cui et al., 1994; Elboutachfaiti et al., 2017; Qian, Cui, Nikiforuk, & Goff, 2012). According to Qian, Cui, Wu, and Goff (2012), the main sugar of the neutral fraction (NF) of the polysaccharides is xylose (68.2%), followed by arabinose (20.2%), galactose (7.9%) and glucose (3.7%), whereas the acidic fraction (AF) is mainly composed by uronic acids (38.7%), containing also rhamnose (38.3%) and galactose (35.2%) in the same proportion. Fucose (14.7%) and xylose (8.9%) are also present but in lower percentages, while arabinose (2.9%) is the less abundant sugar. However, the polysaccharides composition and molecular structures can vary depending on the cultivars/genotype, the environment, the extraction conditions and dehydration process after extraction (Roulard, Petit, Mesnard, & Rhazi, 2016; Ziolkovska, 2012). Thus, when these conditions vary, rheological and other functional properties may be significantly affected (Cui & Mazza, 1996; Cui et al., 1994).

Flaxseed is also a valuable natural source of phenolic compounds, including lignans, phenolic acids, flavonoids, phenylpropanoids, and tannins (Kasote, 2013). There is a vast number of studies reporting that these antioxidant components have pharmacological properties including antidiabetic, antihypertensive, immunomodulatory, anti-inflammatory and neuroprotective properties. The major compounds of

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flaxseed lignans are phytoestrogens (Alu'datt, Rababah, Ereifej, & Alli, 2013; Hao & Beta, 2012; Herchi et al., 2011). These compounds are usually associated to the FS polysaccharides and can be co-extracted during the preparation of the gum, which, although providing antioxidant activity, may cause some setbacks and controversy in food applications due to presence of phytoestrogens as endocrine disruptors (Patisaul & Jefferson, 2010). Therefore, more studies on the phenolic compounds of flaxseed gum and their bioactivity are required.

Thus, in the present work, it is hypothesized that the definition of the extraction conditions of flaxseed gum can be used to tailor its properties according to the envisioned food and pharmaceutical applications. For this, we studied the influence of the extraction temperature on the composition and structural features of flaxseed gum relating such features with the antioxidant capacity and rheological behavior of SFG in aqueous solutions at different pH conditions.

2. Materials and methods

2.1. Materials

Golden flaxseeds were produced in South of Brazil and kindly provided by CISBRA Ltda (Panambi, RS, Brazil). Ethanol was obtained from Dinamica (Brazil); Methanol, DPPH (2,2-diphenyl-1-picrylhydrazyl) and BHA (butylated hydroxyanisole) were purchased from Sigma (USA).

2.2. Extraction of SFG

A physical procedure was used to obtain the polysaccharides with high molecular weight according to Cui, Mazza, Oomah et al. (1994), with some modifications. Firstly, golden flaxseeds were washed with distilled water to remove dirt from the surface. Then, flaxseeds were soaked in distilled water at a flaxseed-to-water concentration of 10% (w/w). This extraction process was made under stirring using an Ultra-Turrax system (IKA RW 20 digital, Brazil) for 5 h at 400 rpm and at three different temperatures (25, 40 and 60 °C). The soaked seeds were filtered (35 Mesh Tyler, Granutest, Brazil) and centrifuged at 11,200 *g* during 10 min. The water containing the dissolved SFG was treated with 99.5% ethanol (1:1) to separate and remove the low molecular weight polysaccharides. Ethanol was then evaporated, and the dialyzed precipitates were freeze-dried (LS3000, Terroni, Brazil).

The SFG yield was determined using the following equation:

$$Yield\ (\%) = \frac{SFG}{Seed} \times 100 \quad (1)$$

where "SFG" represents the total mass of water-soluble portion of flaxseed gum in g (dry weight) after lyophilization and "Seed" represents the mass of flaxseeds used for the extraction in g (dry weight).

2.3. Preparation of SFG aqueous solutions

Aqueous SFG formulations with four concentrations of mucilage (0.75, 1.5, 2.25 and 3% (w/w)) were prepared by dissolving the SFG in deionized water, under magnetic stirring during 3 h at 400 rpm and room temperature. The rheological properties of these formulations were evaluated at pH 3 (0.1 mol L^{-1} citric acid solution) and 6.5 (distilled water).

2.4. Determination of fundamental elemental components and protein content

The fundamental elemental components (carbon, hydrogen, nitrogen and sulfur) were evaluated on a CHNS-O analyzer (Flash 2000, ThermoScientific, UK). Freeze-dried samples of SFG were crushed and homogenized, then weighed into a crucible, closed, and finally placed in the autosampler for instrumental analysis. Protein content was determined using the flaxseed specific factor N × 5.30 for the conversion of nitrogen to SFG protein. All the nitrogen content was considered as protein since the non-protein nitrogen in flaxseed gum is quite low (Qian, Cui, Wu et al., 2012).

2.5. Sugar and Glycosidic-linkage analyses

SFG samples were submitted to a dialysis (12–14 kDa cut-off) in order to obtain the polymeric material. Dialysis was carried out in a walk-in chamber against distilled water at 4 °C under constant stirring during four days, with two water renewals per day. The retentate was centrifuged at 4 °C and 15,000 rpm during 15 min; the supernatant was concentrated, frozen and freeze-dried. Determination of sugars was performed before and after dialysis while linkage analysis was carried out only after dialysis of samples.

Sugars and glycosidic-linkage analysis were performed in order to relate the rheological and antioxidant properties with the structural features of SFG.

2.5.1. Determination of sugars composition

After being submitted to a pre-hydrolysis with 72% H_2SO_4, during 3 h, at room temperature, samples were hydrolyzed with 1 M H_2SO_4 in a heating block, at 100 °C, during 2,5 h. After the first hour, a 500 µl-aliquot was collected from each tube for the analysis of uronic acids, which were determined colorimetrically according to the method referred by Nunes et al. (2012). Galacturonic acid was used as the standard. Total neutral sugars were determined according to the method of Nunes et al. (2012). Briefly, neutral monosaccharides were reduced with $NaBH_4$, acetylated with acetic anhydride in the presence of 1-methylimidazole, and the alditol acetates were extracted with dichloromethane. 2-deoxyglucose was used as the internal standard. After being dissolved in anhydrous acetone, the extracted alditol acetates were analyzed on a GC-FID (PerkinElmer – Clarus 400, Massachusetts, USA) provided with a capillary column DB-225 (30 m length, 0.25 mm internal diameter, 0.15 mm film thickness). The injector temperature was 220 °C and the detector temperature 230 °C. The oven temperature was kept at 220 °C for 7 min; then the temperature increased at 5 °C/min up to 240 °C. Hydrogen was the carrier gas that was injected at 4 bar. Retention times of standards were used to determine and quantify the sugar composition of each of the samples.

2.5.2. Glycosidic-linkage analysis

In order to determine and characterize the glycosidic linkages, the various fractions of the polysaccharides were activated with NaOH pellets after being dispersed in DMSO, according to the method of Ciucanu and Kerek (1984) as indicated by Nunes et al. (2012). After being methylated with CH_3I, each sample was dissolved in $CHCl_3$:MeOH (1:1, v/v), and the solution was dialysed three times against 50% EtOH in distilled water. Then the solution was vacuum dried. Methylated polysaccharides were hydrolysed with 2 M TFA during 1 h in a heating block at 121 °C, vacuum dried, reduced with $NaBD_4$ and acetylated with anhydride acetic in the presence of 1-methylimidazole. The partially methylated alditol acetates were analysed by GC/MS (Shimadzu GC-2010 Plus).

2.6. Zeta potential

Samples extracted at different temperatures were diluted in MilliQ water (Direct-Q3, Millipore, USA) to a concentration of 0.05% (w/w) before being placed in the measuring chamber of microelectrophoresis (Zetasizer Nano-ZS, Malvern Instruments Ltd., UK). Zeta potential was determined as a function of pH, between 2 and 8. The Smoluchowsky model was used to convert the electrophoretic mobility measurement into zeta potential values. The samples were measured in triplicate at 25 °C.

2.7. Antioxidant activity

Radical scavenging activity of the SFG was measured using the DPPH (2,2-diphenyl-1-picrylhydrazyl) method according to Blois (1958), with some modifications. Briefly, 2.5 mL of DPPH (60 μM in methanol) were mixed with 0.2 mL of methanol and 0.3 mL of the sample dissolved in methanol (containing 10 mg mL^{-1}). After vortexing, each solution was stored in the dark for 30 min at room temperature. Then 0.2 mL of each sample was transferred into a Multiskan FC 96-well microplate to measure absorbance at 517 nm (Thermo Scientific, EUA) and the activity was expressed as the percentage of radical scavenging activity (% RSA) relative to the control. All experiments were conducted in triplicate, using the following equation:

$$RSA(\%) = \frac{(Abs_{control} - Abs_{sample})}{(Abs_{control})} \times 100 \quad (2)$$

where Abs_{sample} and $Abs_{control}$ represent the absorbance of the sample solution and the absorbance of the control, respectively. Methanol was used as the control and butylated hydroxyanisole (BHA) was used as the reference antioxidant.

2.8. Total phenolic content (TPC)

TPC was determined using the Folin-Ciocalteu method as described by Wong-Paz et al. (2015). Firstly, the samples were dissolved in distilled water to the concentration of 10 mg/mL (w/v). In order to determine TPC, 800 μL of each sample were mixed with 800 μL of Folin-Ciocalteu reagent (Sigma-Aldrich, USA), shaken and left for 5 min. Then 800 μL of Na_2C0_3 (0.01 M) were added, shaken and left for another 5 min. Finally, the solution was diluted with 5 mL of distilled water and the absorbance was read at 790 nm A calibration curve was prepared using standard solutions of gallic acid (80, 160, 240, 320 and 400 mg/L, $R^2 = 0.9938$). All experiments were performed in triplicate. The TPC was expressed as gallic acid equivalent per 100 g (mg GAE/100 g).

2.9. Phenolic compounds

Freeze-dried SFG samples extracted at different temperatures were analyzed using a Shimatzu Nexpera X2 UHPLC chromatograph equipped with a Diode Array Detector (Shimadzu, SPD-M20 A), according to the methodology used by Sluiter et al. (2008), with some modifications. A 300 mg of each sample were weighted into different pressure tubes and then 3 mL of 72% sulfuric acid were added and mixed with a teflon stir rod for 1 min, until the sample was thoroughly mixed. After that, sample tubes were incubated in water bath for 60 min at 30 °C. Finally, the acid was diluted to a 4% concentration by adding 84 mL of deionized water. Before analysing, the samples were neutralized using calcium carbonate to pH 5-6. Separation was performed on a reversed-phase Aquity UPLC BEH C18 column (2.1 mm × 100 mm, 1.7 μm particle size; from Waters) and a precolumn of the same material, at 40 °C. The flow rate was 0.4 mL min^{-1} with an injected volume of 1 μl. The HPLC grade solvents used were formic acid 0.1% (v/v) in water (up to 100%) as solvent A and acetonitrile as solvent B. The elution gradient for solvent B was as follows: from 0.0 to 5.5 min eluent B at 5%, from 5.5 to 17 min a linear increase to 60%, from 17.0 to 18.5 min a linear increase to 100%; then the column was equilibrated from 18.5 to 30.0 min at 5%. Phenolic compounds were identified comparing their UV spectra and retention times with those of corresponding standards. The various compounds were quantified and identified at different wavelengths: caffeic acid at 320 nm, gallic acid at 280 nm, vanillic acid at 254 nm and ellagic acid at 250 nm.

2.10. Colorimetry analysis

The color of SFG solutions (indicated in the Section 2.3) was measured in triplicate using an Ultra Scan Vis 1043 (Hunter Lab, model Color Quest II, USA) with reflectance mode, CIELab scale L^* (lightness), a^* and b^* (chromaticity parameters), D65 as illuminant and a 10° observer angle as a reference system. Cylindrical coordinates C^* (chroma, represents the intensity) (Eq. (3)) and H^* (hue angle) (Eq. (4)) were calculated from parameters a^* and b^*, according to:

$$C^* = \sqrt{(a^{*2} + b^{*2})} \quad (3)$$

$$H^* = arctan\left(\frac{b^*}{a^*}\right) \quad (4)$$

2.11. Rheological behavior

Flow curves of SFG solutions (from Section 2.3) were obtained using a Physica MCR301 modular compact rheometer (Anton Paar, Graz, Austria) with a stainless-steel plate geometry (75 mm) and 100 μm gap. An up–down–up step program with various shear stresses range for each sample was used to provide shear rate between 0 to 1000 s^{-1} at 25 °C. This wide range includes a number of processes to mimic mastication (Mantovani, Cavallieri, Netto, & Cunha, 2013), and also flowing and mixing. Newtonian (Eq. (5)) and power-law equation (Eq. (6)) were fitted to the data to obtain the rheological properties.

$$\sigma = \eta . \dot{\gamma} \quad (5)$$

$$\sigma = k . \dot{\gamma}^n \quad (6)$$

where σ is the shear stress (Pa), η is the viscosity (Pa.s), k is the consistency index (Pa.s^n), $\dot{\gamma}$ is the shear rate (s^{-1}) and n is the flow index. Eq. (7) was adjusted to the viscosity data according to a power law model in order to evaluate the effect of polysaccharide concentration on viscosity:

$$\eta_{ap}\,(50\,s^{-1}) = K .\, C^B \quad (7)$$

where, η_{ap} (50 s^{-1}) represents the apparent viscosity at a shear rate of 50 s^{-1} (Pa.s), K is a fitting parameter (Pa.s), C is the concentration of SFG (%) and B is the power law exponent (dimensionless) that represents the viscosity dependence with the concentration.

Oscillatory measurements of the SFG solutions (from Section 2.3) were performed using a stress-controlled AR1500ex rheometer (TA Instruments, USA) with a stainless-steel cone-plate geometry (6.0 cm, 2° angle, truncation 67 μm). The viscoelastic properties were evaluated using a frequency sweep between 0.1 and 10 Hz within the linear viscoelasticity domain. These measurements were done at 25 °C after one day of samples storage. The contributions of the elastic and viscous characteristics were evaluated from storage (G') and loss (G'') moduli, respectively.

2.12. Statistical analysis

The data were analyzed using Sigma Plot 11 and Microsoft Excel (Office 365) software. Data were subjected to analysis of variance (ANOVA) ($p < 0.05$) and the means were compared using the Tukey's HSD test to examine if differences between treatments were significant ($\alpha = 0.05$).

3. Results and discussion

3.1. Physicochemical properties

The extraction temperature had a significant impact on the yield, composition and characteristics of SFG. The yield of SFG extraction increased with increasing temperature extraction (Table 1), and the results were in accordance with the ones reported by Cui, Mazza, Oomah et al. (1994) and Kaushik, Dowling, Adhikari, Barrow, and Adhikari (2017), showing an almost two-fold increase when comparing

Table 1
Effect of extraction temperature on the yield, elemental composition and protein content of SFG.

Extraction temperature (°C)	Yield (%, w/w)	N (%, w/w)	C (%, w/w)	H (%, w/w)	Protein content (%, w/w)
25	5.7[c]	0.82 ± 0.01[c]	33.93 ± 0.27[b]	6.14 ± 0.06[a]	4.33 ± 0.07[c]
40	6.9[b]	1.18 ± 0.02[b]	34.83 ± 0.32[a]	6.26 ± 0.09[a]	6.26 ± 0.12[b]
60	10.0[a]	2.60 ± 0.04[a]	35.11 ± 0.54[a]	6.23 ± 0.11[a]	13.80 ± 0.21[a]

[a−c]Different letters in the same column correspond to statistically different samples for a 95% confidence level.

the extraction at 25 °C (5.7% w/w) with the extraction at 60 °C (10% w/w) as shown in Table 3 ($p < 0.05$).

Variations in the concentrations of the various components in plant extracts might be due to the origin, growing conditions and diagenetic alteration of source materials (Fujine, 2008). This means that, although SFG characterization has already been performed (Cui & Mazza, 1996; Cui et al., 1994), such values may not adequately represent the samples used in the present work. Therefore, in order to compare the chemical composition of SFG extracted at different temperatures, samples were evaluated by fundamental elemental composition, zeta potential and total sugars and linkage analyses.

The elemental composition of SFG extracted at different temperatures is presented in Table 1. Nitrogen content increased significantly ($p > 0.05$) with increasing the extraction temperature, which means that the protein content varied between 4.3 ± 0.1% and 13.8 ± 0.2% (w/w), and this is within the range reported by Kaushik et al. (2017). This increase of SFG protein content with the extraction temperature was also observed by Cui et al. (1994), leading to the conclusion that SFG extracted at 60 °C should have better interfacial and emulsifying properties, as demonstrated by Cui and Mazza (1996). Carbon was the major constituent for all the extraction temperatures, indicating the presence of a high content of carbohydrates and some protein in the extracted polysaccharide. Therefore, although an increase in the yield of SFG extraction has been observed, the polysaccharide purity of the extracted decreased with increasing extraction temperature, since the protein yield was also greater at higher temperatures.

The zeta potential of SFG was always negative for pH values between 2 and 8, but a decrease of the absolute value was obtained (Fig. 1) with decreasing pH values. The maximum values of zeta potential for each extraction temperature (25 °C, 40 °C and 60 °C) were −29.37, −34.57 and −35.1 mV, respectively. The isoelectric point (pI) of flaxseed protein isolate is pH 4.2 (Kaushik et al., 2016), but it could not be observed because of the low protein content of the SFG extracted at different temperatures. The lower zeta potential of SFG extracted at higher temperature (or increased anionic character) could be associated to the higher protein content extracted at higher temperatures leading to a more pronounced negative charge at pH above the pI (Kaushik et al., 2017). Indeed, the surface charge became very close to zero at pH near 2, this result is in agreement with Kaushik et al. (2017). At this pH condition, the protein is positively charged and it is near to polysaccharide pKa (Liu, Shim, Shen, Wang, & Reaney, 2017). The maximum negative charge for all SFG samples extracted at different temperatures was observed from pH 6 to 8 and the highest absolute values of surface charge density were observed at higher extraction temperatures, with no difference between 40 °C and 60 °C.

Table 2
Sugar profile and yield of recovering after dialysis of the polysaccharides from flaxseed gum.

SFG	Yield[a] (%, w/w)	mol (%) Rha	Fuc	Ara	Xyl	Gal	Glc	UA	Total sugars (%, w/w)
Before dialysis									
25 °C		8.0	5.1	9.1	34.5	18.5	12.6	12.3	66.5
40 °C		8.0	4.8	9.6	36.2	18.9	6.9	15.6	53.3
60 °C		5.8	2.6	8.2	27.7	16.6	24.3	13.8	47.4
After dialysis									
25 °C	52.9	5.0	4.2	7.5	29.8	13.7	2.5	37.3	63.0
40 °C	60.9	4.6	4.3	8.3	32.9	13.4	2.7	34.8	62.9
60 °C	49.5	4.2	3.9	9.0	30.5	13.4	3.1	35.8	61.5

[a] Yield after dialysis.

The proportion of polymeric material of the samples recovered after dialysis (12–14 kDa cutoff) correspond only to 53%, 61%, and 50% for SFG extracted at 25 °C, 40 °C and 60 °C, respectively (Tables 2 and 3). Nevertheless, after dialysis, the total sugars that composed each sample remained similar for the 25 °C samples (67% and 63% before and after dialysis, respectively), and slightly increased from 53% to 63% for 40 °C samples and from 47% to 62% for 60 °C samples. The sugars composition of the samples determined before and after dialysis allowed also to observe that the main component in all samples was xylose, accounting for about one third in all samples. The dialyzed samples are also rich in uronic acids, accounting also for one third of the carbohydrate's composition, together with 13–14 mol% galactose, 8–9 mol% arabinose, and also rhamnose, fucose, and glucose in amounts ranging from 2 to 5 mol%. As the percentage of rhamnose, galactose and glucose are higher in the raw than in the dialyzed samples, it can be inferred that these carbohydrates are components of low molecular weight polysaccharides. Similar results were reported by Anderson and Lowe (1947) and Cui et al. (1994), Cui, Mazza, Oomah et al. (1994) for the

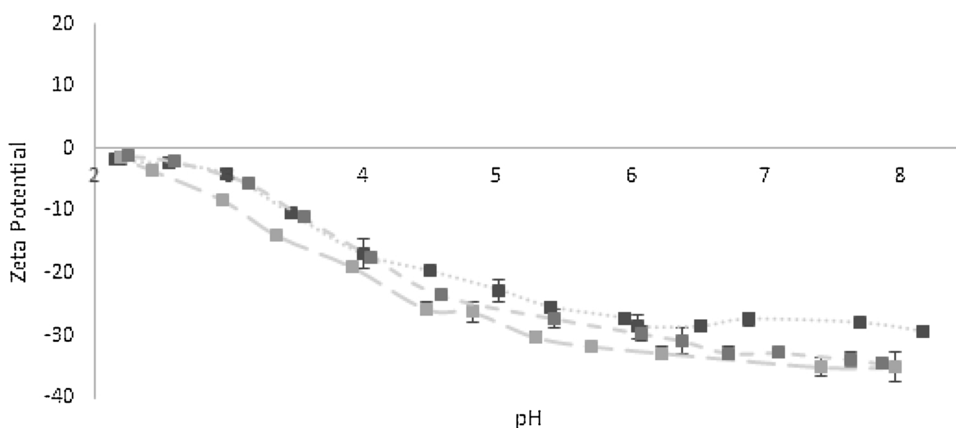

Fig. 1. Zeta potential of SFG aqueous solutions obtained at different extraction temperatures (■ 25 C, ■ 40 C and ■ 60 C). Error bars correspond to a significant difference at $p < 0.05$.

Table 3
Methylation analysis of the polymers extracted from flaxseed gum at three different temperatures, after being dialyzed.

Lincage type	25 °C	40 °C	60 °C
2,3-Rhap	5.0	4.5	8.4
2,4-Rhap	0.4	0.7	0.5
Total Rha	**5.5 (8)**	**5.2 (7)**	**8.9 (7)**
t-Fucp	1.2	3.6	1.9
2,3-Fucp	4.3	4.6	4.8
Total Fuc	**5.5 (7)**	**8.2 (7)**	**6.7 (6)**
2,3-Rhap	5.5 (8)	5.2 (7)	8.9 (7)
t-Araf	2.1	3.5	2.1
3-Araf	0.9	1.2	1.9
5-Araf	5.3	4.0	2.7
2,3,5-Araf	5.1	3.4	5.0
Total Ara	**13.4 (12)**	**12.1 (13)**	**11.8 (14)**
t-Xylp	19.0	14.4	15.5
4-Xylp	17.5	22.4	21.6
2,4-Xylp	1.1	2.9	3.3
3,4-Xylp	0.5	0.9	1.1
2,3,4-Xylp	11.1	9.8	8.8
Total Xyl	**49.2 (47)**	**50.4 (49)**	**50.2 (48)**
t-Gal	8.7	7.1	8.4
4-Galp	6.5	4.2	2.6
6-Galp	2.5	3.3	3.1
3,4-Galp	0.2	0.2	0.2
3,6-Galp	0.3	0.2	0.2
4,6-Galp	1.0	1.6	1.2
2,3,4-Galp	0.4	0.8	0.6
galactitol	0.7	1.2	0.7
Total Gal	**20.4 (22)**	**18.6 (21)**	**17.0 (21)**
t-Glcp	5.7	5.0	5.2
3,4,6-Glcp		0.2	0.1
Total Glc	**5.7 (4)**	**5.2 (4)**	**5.3 (5)**

composition of crude and dialyzed flaxseed gum, except for rhamnose that occurs in lower concentrations in the present study. This seems to be due to the incomplete hydrolysis of the aldobiouronic acid (GalA-Rha) component of the type-I rhamnogalacturonan of flaxseed gum, reported to require at least 6 h at 100 °C at 2 M H_2SO_4 to reach a maximum of release of the rhamnose residues (Emaga, Rabetafka, Blecker, & Paquot, 2012).

The methylation analysis performed to the dialyzed samples allowed to observe that the xylose residues are mainly 1,4-linked (18–22 mol%), representing the unbranched main backbone of the xylan, where proportion of disubstituted 1,2,3,4-Xyl residues accounts for 9–11 mol% and the terminal residues account for 14–19 mol% (Table 3). The relative percentage of the linkages of the xylose residues are quite similar for the higher temperatures of extraction (40 °C and 60 °C), presenting a linear backbone of 1,4-linked xylose, with 22% of unsubstituted residues and 3% of *O*-2 monosubstituted residues and 9–10% of disubstituted residues. At these temperatures of extraction, the relative percentage of disubstituted xylose residues are lower than the one observed for 25 °C, possibly by the higher extractability of the debranched polysaccharides at higher temperature and/or by debranching reactions due to the higher lability of the substituents, thereby increasing the non-substituted units along the 1,4-linked xylose main chain. In these samples it was also quantified arabinose residues, mainly as 1,5-linked, 1,2,3,5-linked, and terminally-linked, which are characteristic of flaxseed arabinoxylan, possibly as substituents at *O*-2 and/or *O*-3 positions of the xylan backbone, together with terminally-linked galactose and xylose residues (Naran, Chen, & Carpita, 2008). The occurrence of 1,2,3-linked rhamnose together with the presence of galactose with a large diversity of linkages, including the terminally-linked, 1,4-Gal, 1,6-Gal, 1,4,6-Gal (Table 3), as well as uronic acids (Table 2), supports the presence of the characteristic homorhamnan domain of the rhamnogalacturonans of flaxseed mucilage (Qian, Cui, Nikiforuk et al., 2012).

These results show that the extracts, although rich in arabinoxylans (Cui et al., 1994; Cui, Mazza, Oomah et al., 1994; Ding, Qian, Goff, Wang, & Cui, 2018; Guilloux, Gaillard, Courtois, Courtois, & Petit, 2009), also have pectic polysaccharides. Nevertheless, the absence of 1,4 and 1,4,6-Glc shows that the xyloglucan reported by Ding et al. (2015), Ding, Cui, Goff, Guo, and Wang (2016) and Ray et al. (2013) is not present in these extracts, as the xyloglucan requires alkali solutions to be extracted.

3.2. SFG phenolic compounds and antioxidant capacity

SFG samples extracted at 60 °C showed higher ($p < 0.05$) antioxidant activity than samples extracted at lower extraction temperatures (Table 4). Previous works have shown the relationship between antioxidant activity and the concentration of phenolic compounds in plants, correlating several analyses of antioxidant capacity, for example, ORAC (Oxygen Radical Absorbance Capacity), TRAP (Total Radical-trapping Antioxidant Parameter), DPPH (2,2-diphenyl-1-picrylhydrazyl method), ABTS (2,2′-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) and HORAC (Hydroxyl Radical Antioxidant Capacity), with the phenolic content. After those studies, the researchers observed that an increase in phenolic content leads to an increase of the antioxidant activity (Číž et al., 2010; Rajurkar & Hande, 2011; Seçzyk, Swieca, Dziki, Anders, & Gawlik-Dziki, 2017). In addition, Hao and Beta (2012) observed that the antioxidant activity of flaxseed hull exhibited a large variation between different varieties, with IC_{50} values ranging between 4.95–8.23 g L^{-1} of phenolic compounds. Further, the antioxidant activity of free phenolic compounds extracted from the full fat flaxseed under heating was higher (62.3%) when compared to the free phenolic compounds extracted without heat treatment (44.0%) (Alu'datt et al., 2016).

A number of studies using similar analyses have shown that the total phenolic content could be used as an indicator of antioxidant activity (Abozed, El-kalyoubi, Abdelrashid, & Salama, 2014; Oliveira et al., 2012; Piluzza & Bullitta, 2011) although the total phenolic content does

Table 4
Effect of the extraction temperature on phenolic compounds profile and antioxidant activity of SFG.

Extraction temperature	25 °C	40 °C	60 °C
Antioxidant activity (% RSA)*	4.39 ± 1.52[c]	12.27 ± 2.87[b]	29.64 ± 2.39[a]
TPC (mg GAE. 100 g^{-1})*	12.37 ± 0.59[b]	13.01 ± 0.20[b]	18.60 ± 0.08[a]
Phenolic compound (mg L^{-1})			
Caffeic acid	6.58 ± 0.06[a]	6.39 ± 0.02[b]	6.06 ± 0.11[c]
p-cumaric-acid + epicatechin	1.60 ± 0.14[a]	1.43 ± 0.00[b]	1.43 ± 0.00[b]
Ellagic acid	1.18 ± 0.54[a]	1.05 ± 0.07[a]	3.14 ± 0.46[b]
Cinnamic acid	2.28 ± 0.02[a]	2.26 ± 0.01[a]	2.27 ± 0.01[a]
Vanillic acid	–	–	5.42 ± 0.00

[a−c]Different letters in the same line correspond to statistically different samples for a 95% confidence level.
* Sample at 10 mg/mL.

not incorporate all the antioxidants. Moreover, the structure of the antioxidants and the interactions between them should also be considered. Therefore, it is reasonable to consider that the antioxidant activity of the extracts can be related with the presence of some individual active phenolic compounds and their synergism in the mixture (Piluzza & Bullitta, 2011). In the present work, it was verified that the increase in antioxidant activity was directly related to with the content of phenolic compounds.

In this study, the total phenolic content was estimated from the reaction between the Folin-Ciocalteu reagent and phenolic benzene rings. It was observed that the TPC of SFG was significantly influenced by the extraction temperature: the SFG extracted at 60 °C showed the highest ($p < 0.05$) TPC values, followed by those extracted at 40 and 25 °C (Table 4). Due to the affinity between the phenolic compounds and the protein bonds, it is probable that some phenolic compounds have been extracted in greater quantity at higher temperatures by dragging since the increase of the extraction temperature also increased the protein content. These results compare well with those reported for guar gum (15.0 mg GAE. 100 g^{-1}) and are half of those reported for locust bean gum (33.0 mg GAE. 100 g^{-1}) (Hamdani & Wani, 2017).

In order to confirm the antioxidant activity and relate with the TPC profile, phenolic compounds present in the various SFG samples were determined by UHPLC (Table 4).

The quantitative and qualitative composition of phenolic compounds in extracted SFG was dependent on the extraction temperature, although more obvious for 60 °C and mainly for ellagic and vanillic acids. Caffeic acid and p-cumaric-acid + epicatechin concentrations decreased and ellagic acid concentration increased with increasing extraction temperature. The amount of extracted phenolic compounds detectable by the methodology used was higher for extraction at 60 °C (18.32 mg.L^{-1}) followed by extraction at 25 °C (11.64 mg.L^{-1}) and 40 °C (11.13 mg.L^{-1}). These results are in agreement with the TPC values, however it was observed that the antioxidant activity measured in the SFG extracted at 40 °C was about three times greater than at 25 °C. This fact may be due to a) other extracted non-phenolic compounds which also exhibit antioxidant activity (extracted dry matter at 40 °C was higher when compared with extraction at 25 °C, as can be observed in Table 1) or b) a high number of interactions between phenolic compounds and proteins extracted in greater quantity at higher temperatures, making difficult to identify a given compound as a phenolic compound.

Almeida, Cavalcante, and Vicentini (2016) studied the cytotoxicity, antiproliferative activity, and protection from DNA-induced damage in HTC cells, showing that vanillic acid was effective at protecting DNA from damage at any concentration between 1.684 mg.L^{-1} and 16.84 mg.L^{-1}. In this study, the vanillic acid values detected were 5.42 mg.L^{-1}, which is in the range studied by the mentioned authors. The affinity of phenolic compounds to conjugate with major food components such as proteins, carbohydrates, lipids and minerals is due to the presence of an aromatic ring with hydroxyl groups and carboxylic acids, which is the case of vanillic acid; in this case, such affinity may have been the cause of the presence of this phenolic compound in SFG extracted at 60 °C (Alu'datt et al., 2016; Sabally, 2006). Lutz, Lugli, Bitsch, Schlatter, and Lutz (1997) studied the dose-response effect of different caffeic acid concentrations in rats, concluding that this compound can present anti-tumor properties in concentrations above 0.05%. They also claim that, according to data collected, concentrations above 2% of caffeic acid may have anticarcinogenic properties. Since for all extraction temperatures, the concentration of caffeic acid is approximately 0.0006%, it can be concluded that the dose of SFG to be consumed by rats should be high to observe some therapeutical effect. Regarding ellagic acid, previous studies have shown that even at very low concentrations this compound has a high antioxidant activity (Festa et al., 2001; Han, Lee, & Kim, 2006; Kilic, Yeşiloğlu, & Bayrak, 2014). Further, Priyadarsini, Khopde, Kumar, and Mohan (2002) demonstrated that the ellagic acid concentration required to inhibit 50% of lipid peroxidation was about 0.95 mg.L^{-1}, which is lower than the amount present in SFG (between 1.05–3.14 m.L^{-1}). Cinnamic acid was also detected and its concentration kept constant regardless the extraction temperature. Vanillic acid was only detected for SFG extracted at 60 °C; perhaps the extraction of this compound is also enhanced with temperature and the amount extracted at lower temperatures kept below the detection limits of the method. Similar results were observed by Sytar, Hemmerich, Zivcak, Rauh, and Brestic (2018), who studied the composition of 26 medicinal plants. All of them showed high antioxidant activity, and vanillic acid was present as the major phenolic compound in some of them (extraction temperatures above 60 °C were used for these analyzes).

These compounds have been widely studied, since they provide protection e.g. from the deleterious effects of oxidative stress (Cremonini, Bettaieb, Haj, Fraga, & Oteiza, 2016). While anti-oxidant effects are the most studied in the literature, this being both a consequence and a motivation for the very extensive amount of work reported so far, it is also true that many other biological activities have been identified and demonstrated. For example, ellagic acid (and its dimeric derivative) also exhibits anti-mutagenic, anti-carcinogenic and anti-inflammatory activity (Feng et al., 2009), and caffeic acid shows anti-dementia properties, contributing to reduce the progression of neuronal degenerations such as Alzheimer's disease (Akomolafea et al., 2017; Mallik et al., 2016). Further, cinnamic acid and its derivatives have attracted attention due to their anticarcinogenic, antimicrobial, antidiabetic, anticonvulsant, antidepressant, neuroprotective, analgesic, anti-inflammatory, muscle relaxant and sedative properties (Oishia, Yamamotoa, Oikea, Ohkurae, & Taniguchif, 2017). Furthermore, vanillic acid has been associated with a variety of pharmacological activities, such as anti-carcinogenic, anti-apoptotic and anti-inflammatory but it has become most popular for its pleasant creamy odor that is widely used in fragrances, and licensed as a food additive, due to its distinct vanilla flavor (Gitzinger et al., 2012; Vinothiya & Ashokkumar, 2017). This acid has also shown to reduce the action of amylase, the primary human carbohydrase enzyme, thus reducing the efficiency of the digestive process in the mouth (Dupuis, Tsao, Yada, & Liu, 2017).

The significant changes observed in the SFG composition upon extraction at different temperatures, particularly those regarding phenolic compounds; both their qualitative and quantitative compositions lead us to believe that it is possible to tailor to some extent the bioactive/functional properties of SFG extracts by controlling the extraction temperature.

3.3. Colorimetric analyses

Color is a crucial parameter with a significant role in the acceptability of foods. Reflectance spectrophotometry results indicated a change in the color of the samples mainly due to a significant ($p < 0.05$) increase of the lightness parameter (L^*) when decreasing SFG extraction temperature (Table 5). This may be important when using SFG as food ingredient, depending on SFG concentration used in the final food formulation. Samples extracted at 60 °C also showed a higher chroma (C^*) value, which is associated with color saturation and tended to increase with SFG concentration. This is possibly due to the increase in phenolic compounds content in the SFG extract (as shown in Section 3.2), to the higher concentration of proteins (as reported in Section 3.1) or to the occurrence of Maillard reaction (especially relevant at 60 °C, considering the 5 h of extraction time). The Hue angle (all below 2°), H^*, a measure of color intensity, was located in the first quadrant, between yellow and red, and showed no significant differences between 40 and 60 °C extraction temperature. The obtained values for H^* were higher for samples extracted at 25 °C; the same happened for the value of L*, which means that the addition of SFG extracted at lower temperatures will exert less influence on the color of food products.

Table 5
L^*, C^* and H^* values of SFG solutions obtained at different extraction temperatures (25 °C, 40 °C and 60 °C) and prepared with varied SFG concentrations (0.75%, 1.5%, 2.25% and 3% w/w).

	SFG (%)		L*	C*	H*
Extraction Temperature	**25 °C**	**0.75**	87.0 ± 0.10[a]	1.0 ± 0.10[h]	1.1 ± 0.02[b]
		1.5	85.2 ± 0.10[c]	0.76 ± 0.10[j]	1.3 ± 0.11[a]
		2.25	85.5 ± 0.20[c]	1.15 ± 0.12[g]	1.4 ± 0.12[a]
		3	85.4 ± 0.01[c]	1.29 ± 0.03[f]	1.4 ± 0.01[a]
	40 °C	**0.75**	85.9 ± 0.11[b]	0.91 ± 0.02[h]	0.7 ± 0.12[c]
		1.5	85.8 ± 0.01[b]	2.14 ± 0.14[e]	0.8 ± 0.01[c]
		2.25	85.6 ± 0.10[b]	2.57 ± 0.02[d]	0.8 ± 0.02[c]
		3	85.5 ± 0.10[b,c]	3.18 ± 0.01[c]	0.8 ± 0.02[c]
	60 °C	**0.75**	80.1 ± 0.10[e]	1.8 ± 0.21[e]	0.7 ± 0.13[c]
		1.5	80.9 ± 0.02[d]	3.3 ± 0.10[b]	0.7 ± 0.04[c]
		2.25	81.2 ± 0.22[d]	5.5 ± 0.03[a]	0.8 ± 0.01[c]
		3	81.1 ± 0.23[d]	5.4 ± 0.14[a]	0.8 ± 0.01[c]

[a−h]Different letters in the same column correspond to statistically different samples for a 95% confidence level.

3.4. Rheological properties

Rheological properties of SFG solutions can be significantly affected by variables such as shear rate and time, pH and extraction temperature of the polysaccharide (Cui, Mazza, Oomah et al., 1994; Kaushik et al., 2017), which will be influenced by the concentration of polysaccharide needed to achieve the desired viscosity or other rheological characteristic. Thus, the study of rheological properties was performed to evaluate thickening properties and viscoelastic behavior of SFG aqueous solution. The influence of pH and SFG concentration on rheological properties was assessed under isothermal conditions.

3.4.1. Flow curves analysis

All samples presented a shear time-independent behavior (Suplementary material). The arbitrarily positioned chains of polymer molecules when the fluid is at rest become aligned in the same direction of the flow as shear rate increases, decreasing the solution viscosity (Koocheki, Reza-Taherian, & Bostan, 2013). A similar behavior was observed for dispersions of chia (*Salvia hispanica* L.) mucilage (Capitani, Ixtaina, Nolasco, & Tomás, 2012), *Opuntia ficus indica* (Medina-Torres, Brito-De La Fuente, Torrestiana-Sanchez, & Katthain, 2000), *Lepidium sativum* (Karazhiyan et al., 2009), tragacanth (Chenlo, Moreira, & Silva, 2010) and *Lepidium perfoliatum* (Koocheki et al., 2013) gums. The effects of the extraction temperature, SFG concentration and pH on flow curves are indicated in Table 6. SFG solutions showed a shear-thinning behavior, as n values were lower than 1 regardless of the extraction temperature.

Apparent viscosity values at a fixed shear rate of 50 s^{-1} ($\eta_{ap\ 50}$) for the different extraction conditions are shown in Fig. 2. This shear rate value was chosen as representative of the food mastication (Wood, 1968). Increasing SFG concentration caused an increase in the apparent viscosity of the solutions (Table 6), possibly due to the higher content of total solids in the solution, hindering the intermolecular movement induced by hydrodynamic forces (Capitani et al., 2015). The increase of the extraction temperature decreased the apparent viscosity and pseudoplasticity of the SFG solutions, which can be directly related to: a) an increase of protein content and b) interaction between polysaccharide chains and proteins (mixed and discontinuous network) (Fedeniuk & Biliaderis, 1994; Qian, Cui, Wu et al., 2012). Apparent viscosity values tended to decrease with decreasing pH values mainly at low SFG concentrations, which could be related to the lower magnitude of surface charge density (or solubility) when compared to SFG solutions at pH 6.5 (Fig. 1). Thus, the decrease in viscosity could be attributed to a lower repulsion between SFG compounds (Hosseini, Reza, Mozafari, Hojjatoleslamy, & Rousta, 2017).

In addition, data of apparent viscosity (at 50 s^{-1}) for different concentrations of flaxseed gum and obtained at different extraction temperatures were adjusted by Eq. (7). Results presented in Table 7 confirm that the increase of the SFG extraction temperature caused a decrease in the viscous behavior of the aqueous solutions (decrease of K value). The effect of SFG concentration on the viscosity of aqueous FG solutions (B value) at different pH values was also affected by the extraction temperature ($p < 0.05$). At pH 3 a significant decrease of the power law exponent (B value) of the SFG extracted at the highest temperature means that in this condition the increase of SFG concentration exerted a minor effect on the viscosity. This behavior can be associated to a higher amount of some compounds extracted at 60 °C, such as phenolic compounds and proteins, and possibly to the reduction of substituents in the xylose chains (reported in Table 3), which may contribute to a lesser extent of interchain bonds and thus to a lower

Table 6
Steady state rheological properties of aqueous solutions of SFG obtained at different extraction temperatures (25 °C, 40 °C and 60 °C). SFG solutions were prepared at various concentrations and pH values. Rheological measurements were obtained in triplicate at 25 °C.

	SFG (%)		pH 6.5		pH 3	
			n	k (Pa s^n)	n	k (Pa s^n)
Extraction Temperature	**25 (°C)**	0.75	0.83 ± 0.00[b]	0.05 ± 0.00[g]	0.86 ± 0.01[b]	0.03 ± 0.00[h]
		1.5	0.72 ± 0.01[c]	0.28 ± 0.01[f]	0.70 ± 0.01[c]	0.26 ± 0.00[f]
		2.25	0.60 ± 0.00[e]	1.56 ± 0.03[c]	0.61 ± 0.04[d,e]	1.43 ± 0.00[c]
		3	0.58 ± 0.01[f]	3.12 ± 0.08[a]	0.56 ± 0.01[e]	3.06 ± 0.00[a]
	40 (°C)	0.75	0.83 ± 0.00[b]	0.05 ± 0.00[g]	0.87 ± 0.01[b]	0.03 ± 0.00[h]
		1.5	0.71 ± 0.00[c]	0.28 ± 0.00[f]	0.81 ± 0.09[b]	0.31 ± 0.10[e,f]
		2.25	0.67 ± 0.03[d]	0.63 ± 0.50[d]	0.64 ± 0.00[d]	0.62 ± 0.01[d]
		3	0.56 ± 0.02[f]	2.66 ± 0.02[b]	0.59 ± 0.03[e]	2.09 ± 0.05[b]
	60 (°C)	0.75	0.93 ± 0.00[a]	0.01 ± 0.00[h]	0.99 ± 0.03[a]	0.01 ± 0.00[i]
		1.5	0.83 ± 0.01[b]	0.06 ± 0.00[g]	0.87 ± 0.01[b]	0.04 ± 0.01[h]
		2.25	0.73 ± 0.02[c]	0.27 ± 0.01[f]	0.84 ± 0.02[b]	0.08 ± 0.01[g]
		3	0.73 ± 0.03[c]	0.55 ± 0.00[e]	0.72 ± 0.01[c]	0.36 ± 0.00[e]

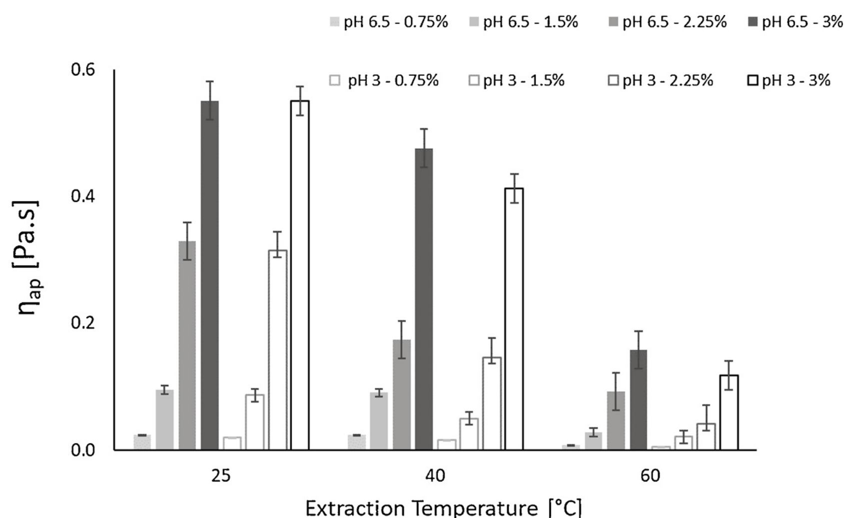

Fig. 2. Apparent viscosity of SFG solutions obtained at different extraction temperatures (25 °C, 40 °C and 60 °C) and pH values, at a fixed shear rate of 50 s^{-1}. Full and empty symbols correspond to solutions at pH 6.5 and 3, respectively.

viscosity. This behavior was not observed at pH 6.5, since parameter *B* remained practically constant for all the extraction temperatures, indicating that the phenolic compounds and proteins are more sensible to acid pH values.

3.4.2. Oscillatory analysis

The effects of the extraction temperature, concentration and pH of SFG solutions on viscoelastic properties are shown in Fig. 3A (pH 6.5) and B (pH 3). For all concentrations, regardless the pH of the SFG solution and the extraction conditions, the samples presented a predominance of viscous properties, the liquid-like properties predominate over that of solid-like, in the frequency range of 0.1–10 Hz, as the viscous modulus (*G”*) was always greater than the elastic modulus (*G’*). Although SFG solutions exhibit mainly a viscous behavior, previous works have shown that when this xylan is dissolved together with another hydrocolloid, both *G’* and *G”* increased indicating the formation of a stronger network. For example, Li et al. (2012) obtained higher values of *G’* and *G”* for solutions with 15% of casein plus 0.5% SFG when compared to solutions with 19% of casein, as well as showing that solutions with 15% of casein had lesser *G’* and *G”* than solutions with 15% of casein plus 0.5% of SFG. This is possibly due to the establishment of interactions between the polymer chains of the different gums, pointing to the possibility of improved tailoring of SFG’s textural properties through combinations with other hydrocolloids.

Higher *G′* and *G”* values were observed for SFG extracted at lower temperatures and these results are in agreement to the trend observed by Cui, Mazza, Oomah et al. (1994). Similar tendency was observed for higher SFG concentrations. Therefore, the lowest values of *G’* and *G”* were observed for the sample with the lowest amount of SFG (0.75% w/w) extracted at 60 °C. In addition, *G’* and *G”* were more frequency-dependent for an extraction temperature of 60 °C, which can be associated with the formation of a less complex structure (again, possibly due to the lower amount of interactions, as a consequence of the reduction of substituents in the xylose chains – see Table 3) as also observed by Cui, Kenaschuk, and Mazza (1996). Polysaccharides extracted from yellow flaxseeds presented higher *G′* and *G″* properties and apparent viscosity at higher levels of xylose followed by arabinose and galactose (Cui et al., 1994; Cui, Mazza, Oomah et al., 1994). Therefore, one of the factors that may have influenced the increase of rheological properties is the high xylose and arabinose content observed before dialysis (Table 2). However, SFG extracted at 40 °C and 60 °C presented weaker rheological properties, despite of the xylose and arabinose content of SFG extracted at 40 °C was similar to 25 °C. These results could be associated to the decrease of water absorption capacity (WAC) of SFG with the increase of the extraction temperature, since polysaccharide granules can be not properly swollen at 40 °C. SFG extracted at lower temperatures showed higher pseudoplastic character, which can be associated to flaxseed granules significantly swollen, leading to a more visible deformation under shear forces. As observed for the apparent viscosity values (Fig. 2), the decrease of pH exerted a negative effect on the rheological properties, being this effect more pronounced at lower concentrations (Fig. 3A and B).

4. Conclusions

The extraction temperature affected SFG composition and physical properties (rheology and color). In particular, it was shown that the composition in phenolic compounds (caffeic acid, p-cumaric-acid + epicatechin, ellagic acid, cinnamic acid and vanillic acid were identified and quantified) was affected by the extraction temperature, which might have influenced the antioxidant capacity of the samples. Given that the concentrations of different phenolic compounds were affected differently for each of the extraction temperatures, it is hypothesized that the resulting SFG extracts may have diverse bioactive/functional properties.

The rheological properties at low and high deformations were

Table 7
Fitting parameters obtained from the power law equation relating viscosity at 50 s^{-1} and SFG concentration. SFG was extracted at different extraction temperatures (25 °C, 40 °C and 60 °C) and aqueous solutions were prepared at varied pH (3 and 6.5).

pH	25 °C			40 °C			60 °C		
	K	B	r^2	K	B	r^2	K	B	r^2
3	0.0341^a	2.550^a	0.9922	0.0382^a	2.124^b	0.9386	0.0139^b	1.712^c	0.9454
6.5	0.0458^a	2.250^a	0.9886	0.0424^a	2.013^b	0.9770	0.0139^b	2.334^a	0.9949

[a–c]Different letters in the same line for each parameter correspond to statistically different samples for a 95% confidence level.

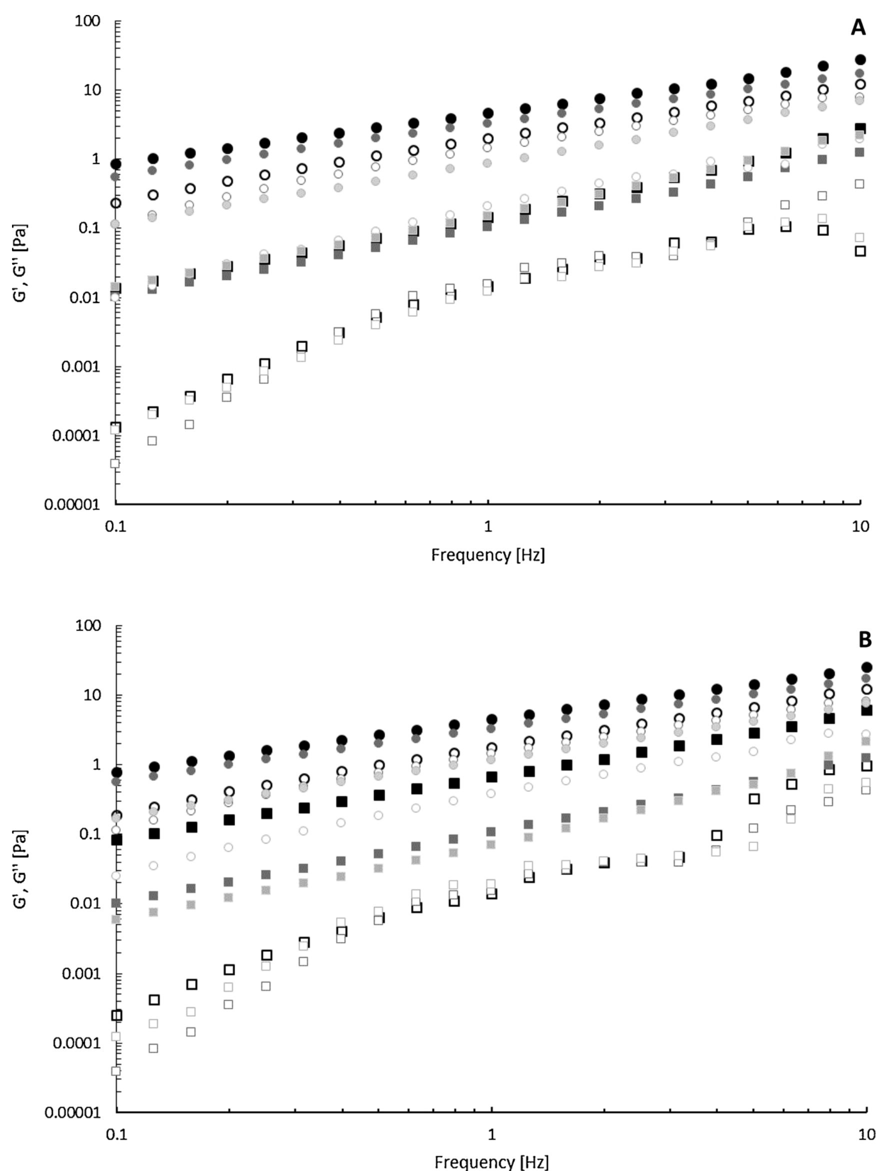

Fig. 3. Elastic modulus (open symbols) and viscous modulus (closed symbols) as a function of frequency under isothermal (25 °C) conditions for solutions of SFG extracted at 25 °C (black symbols), 40 °C (dark grey symbols) and 60 °C (light grey symbols) in pH 6.5 (A) and pH 3 (B) for the extreme concentrations studied: (■) 0.75% and (●) 3% (w/w).

negatively affected by the increase of the extraction temperature. Such a behavior can be related to the protein content increase, the reduction of substituents in the xylose chains, and/or the interaction between protein and polysaccharide molecules.

Overall, the extraction temperature affected both the biological/functional activities and the rheological properties of SFG: viscous properties decreased with increasing extraction temperature and phenolic composition changes leading to a higher antioxidant capacity.'

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.carbpol.2019.02.078.

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