# Properties of mucilage blends using psyllium husk (*Plantago psyllium* L) and chia seed (*Salvia hispanica* L)

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## ABSTRACT

The food industries face a constant challenge to use fewer and fewer ingredients in food composition and to increase the sustainability and nutritional value of these ingredients. The interaction and efficiency of gels can be enhanced by using polysaccharide mixtures. As an alternative for using gels as thickeners and stabilizers, this study used mucilage mixtures of psyllium (P) and chia (C) obtained by blending or combined extraction to investigate the interaction properties of these potential polysaccharide ingredients. After the extraction of psyllium husk and chia seed, separately, and a third extraction combining the two sources simultaneously, six samples were prepared with the following percentages: 100% P, 75-25% P-C, 50%-50% P-C, 25%-75% P-C and 100% C, named T1 to T5. The combined mucilage extraction was called T6. Factors such as pH, oBrix, and oil-holding capacity revealed no significant difference between the samples; higher carbohydrates values were indicated for the content of chia seed (13.14 g/L). Water solubility ranged from 7 to 51.25% without the occurrence of an interaction effect. The thermal effects were similar to natural hydrogels and the chia mucilage revealed less weight loss during the major breakdown stage of decomposition. The corroboration of the interaction property occurred through the viscosity factor. The viscosity of the combined sample (T6) had higher values than the other samples and the attenuated total reflection (ATR) spectra indicated more molecule conformation similarities with the psyllium than chia.

Keywords: Gels. Hydrocolloid. Stabilizer. Thermal analyses (TGA/DSC). Thickener.

## RESUMO

*As indústrias de alimentos enfrentam um desafio constante de utilizar cada vez menos ingredientes na composição dos alimentos e de aumentar a sustentabilidade e o valor nutricional destes ingredientes. A interação e eficiência de géis podem ser reforçadas com a utilização de misturas de polissacarídeos. Como alternativa à utilização de géis como espessantes e estabilizantes, este estudo utilizou misturas de mucilagens de psyllium (P) e chia (C) obtidas por mistura ou extração combinada para investigar as propriedades de interação destes potenciais ingredientes polissacarídeos. Após a extração da casca de psyllium e da semente de chia, separadamente, e uma terceira extração combinando as duas fontes simultaneamente, foram preparadas seis amostras com as seguintes percentagens: 100% P, 75-25% P-C, 50%-50% P-C, 25%-75% P-C e 100% C, denominados T1 a T5. A extração combinada das mucilagens chamou-se T6. Fatores como o pH, °Brix, e a capacidade de retenção de óleo não revelaram diferenças significativas entre as amostras; foram indicados valores mais elevados de carboidratos para o conteúdo de sementes de chia (13.14 g/L). As solubilidades em água variaram de 7 a 51.25% sem a ocorrência de um efeito de interação. Os efeitos térmicos foram semelhantes aos hidrogéis naturais e a mucilagem da chia revelou menor perda de massa durante a maior fase de decomposição. A corroboração da propriedade de interação ocorreu através do fator de viscosidade. A viscosidade da amostra combinada (T6) teve valores mais elevados do que as outras amostras e os espectros de reflexão total atenuada (ATR) indicaram mais semelhanças de conformação da molécula com o psyllium do que o chia.*

Palavras-chave: *Géis. Hidrocolóide. Estabilizante. Análises térmicas (TGA/DSC). Espessante.*

**SUBMETIDO** 03/11/2019 > **APROVADO** 22/09/2020

## 1 Introduction

Hydrocolloids are widely used by the food industries to ensure desirable textures to processed foods, retaining water and representing notable thickeners and stabilizers (CASAS, 2016; MUÑOZ *et al.*, 2012). Several types of hydrocolloids are used, which may be originated from plant exudates such as gums and starches, as well as microbial and synthetic ones (GOFF; GUO, 2019; CASAS, 2016). Hydrocolloids are also often referred to gums or mucilage (GOFF; GUO, 2019).

Certain hydrocolloids present disadvantages, for instance, starches can contribute significantly to the caloric content of food (CASAS, 2016). In addition, both the demand for sustainable ingredients with the highest nutritional potential and the so-called "clean label" foods (with as few ingredients as possible) are growing and becoming challenges for the industries (GOFF; GUO, 2019; SOUKOULIS; GAIANI; HOFFMANN, 2018). As a proposal to add more healthiness to processed foods, seed mucilage and plant husk can be used, which besides presenting good gel formation potential, are also associated with benefits such as regulation of intestinal microflora, blood lipid levels, antioxidant and anti-inflammatory activity, among others (SOUKOULIS; GAIANI; HOFFMANN, 2018; FERNANDES; SALAS-MELLADO, 2017).

Psyllium, found in plant seeds of the genus *Plantago*, contains soluble fibers that are functional hydrocolloids (GUO *et al.*, 2009, RAHAIE *et al.*, 2012). The husk is the main product of psyllium and methods for its extraction are known (GUO *et al.*, 2009). Considered a low-cost polysaccharide source of great technological potential, it is renewable and presents biodegradability characteristics and hydro affinity that allow its application as hydrogel (THAKUR; THAKUR, 2014).

Another potentially functional food is chia, which originates from annual herbaceous plants belonging to the *Lamiaceae* family. The immersion of chia seeds in water exudes its hard-adhered mucilage (FERNANDES; SALAS-MELLADO, 2017; CAPITANI *et al.*, 2012). No references were found for extraction of chia seeds husk. This mucilage has physicochemical properties responsible for its rising as an ingredient for the production of bread, cakes and desserts, substituting greasy ingredients (BORNEO; AGUIRRE; LEON, 2010, CAPITANI *et al.*, 2012).

The interaction and efficiency of gels can be enhanced by using polysaccharide mixtures, called mixed gels, such as in diluted solutions of xanthan gum and locust beans, which only present significant shear stress when combined (WALSTRA; VLIET, 2010). Studies are needed to improve the productivity of hydrocolloid sources and research evaluating the synergy between different sources is also required (YEMENICIOGLU *et al.*, 2019; CASAS, 2016).

> This study is an investigation of the interaction between two emerging thickening ingredients: psyllium husk and chia seed mucilage. Aqueous extraction was applied independently in the psyllium and chia mucilage, as well as in the mixture of different proportions of psyllium and chia mucilage, in order to evaluate their characteristics and study their synergy.

## 2 Material and methods

### 2.1 Material

Psyllium husk (P) and chia seed (C) from the respectively species *Plantago psyllium* L and *Salvia hispanica* L, were obtained in a local market and sent to Cereals Technology laboratory in the State University of Maringá. The husk and seed were stored separately in polyethylene bags under normal conditions of brightness and room temperature until the extraction process.

### 2.2 Mucilage extraction

Chia mucilage extraction was carried out based on Muñoz *et al.* (2012) by inserting whole seeds in a beaker with distilled water 1:40 (w/v) at 80 °C. After a two-hour constant stirring using a screw propeller stirrer, the aqueous suspension extracted was filtered in cloth, spread on a drying tray and air-forced dried at the temperature of 50 °C during 20 hours. The mucilage was subjected to hermetic storage and refrigeration (about 5 °C). Ratio of 1:100 (w/v) was established for the psyllium mucilage extraction based on Ahmadi *et al.* (2012), using distilled water at 80 °C during one-hour stirring and subjected the aqueous suspension to a double filtering in cloth with the same drying and storage conditions employed for the chia mucilage. The third extraction consisted of a combination of the above-mentioned methods. Considering the use of different types of raw materials (husk and seed) and considering the psyllium swelling index being higher than that of chia, 50 g of chia seed was added into a beaker with distilled water at 80 °C (1:40 w/v) and after one-hour stirring, 20 g of psyllium husk; the extraction continued for another hour and the aqueous suspension was subjected to double filtered in cloth. The drying and storage conditions remained the same employed to the previous samples. Mucilages extraction efficiencies were measured by dividing the weight of the mucilage obtained through the extraction by the initial raw material weight and multiplied by 100. Equation (1) was used to estimate the combined mucilage efficency ($e_{CM}$).

$$e_{CM}\,(\%) = \frac{m_P \times e_P + m_C \times e_C}{m_P + m_C} \times 100 \tag{1}$$

Where mP and mC are the weight of psyllium and chia, respectively, eP and eC are the extraction efficiency of the isolated extraction previously carried out of psyllium and chia, respectively.

### 2.3 Blends preparation

The present study was conducted with six treatments using mucilage powders of psyllium husk (P) and chia seed (C). The mixing range was: (T1) 100% P; (T2) 75% P, 25% C; (T3) 50% P, 50% C; (T4) 25% P, 75% C and (T5) 100% C. The combined mucilage was denominated T6. For a complete homogenization, each treatment was dissolved in distilled water 1:100 (w/v), scattered using a mixer, and lyophilized (freeze-dried) for 48 hours.

The reconstitution of freeze-dried mucilages was based on the León-Martínez, Méndez-Lagunas and Rodríguez-Ramírez (2010) method, with modifications. A magnetic stirrer (Fisatom 7BD) was used to scatter the mucilage (room temperature, 90 min) and distilled water to prepare the 1:100 (w/v) rate solutions.

### 2.4 Total carbohydrates content, soluble solids and pH

The content of total carbohydrate of the reconstituted mucilages was assessed through the phenol-sulfuric method described by Dubois *et al.* (1956). A refractometer Briobrix was used to determine the soluble solids content (ºBrix). The pH was determined using potentiometric measurement at 25 ºC.

### 2.5 Oil-holding capacity (OHC)

OHC was assessed based on Capitani *et al.* (2012) by subjecting an aliquot of 10 mL of one percent reconstituted mucilages to a two-minute homogenization using a Fisatom 7BD magnetic stirrer at 5000 rpm. An oil-in-water emulsion was prepared by adding 10 mL of soybean oil under a constant three minutes stirring. The emulsion was subjected to a 30-minute centrifugation at 455 × g; subsequently, the measure of the supernatant oil volume was performed. Oil-holding capacity was expressed as mL oil held per mL sample.

### 2.6 Water solubility index (WSI)

Samples of the freeze-dried mucilages powder were dissolved in 30 mL of distilled water (1:100 w/v), scattered using a magnetic stirrer (Fisatom 7BD) for five minutes at 60 ºC and centrifuged at 455 × g for 30 minutes (20 ºC). The supernatant was subjected to a six-hour drying process at 100 ºC and the final weight was divided by the initial weight with the result multiplied by 100, indicating the value for the water solubility index.

### 2.7 Thermal characteristics

TGA (Thermogravimetric analysis) and DSC (Differential Scanning Calorimetry) were performed in freeze-dried samples according to Iqbal *et al.* (2011b) using a simultaneous thermal analyzer NETZSCH STA 409 PG/PC, under nitrogen atmosphere at a flow rate of 50 $cm^3 min^{-1}$ with temperature ranging from 25 ˚C (room temperature) to 600˚C, at a rate of 10 ˚C $min^{-1}$, using platinum crucible. TGA peak was found through differentiate TGA curve.

### 2.8 Viscosity

A controlled stress rheometer (Brookfield DV-III, USA) with a concentric cylinder geometry (SC4-27) was used for the viscosity analysis of the freeze-dried reconstituted mucilage. The constant temperature of 25 ºC was maintained using a water bath (Brookfield TC-502, USA).

### 2.9 Attenuated total reflection (ATR) spectra

ATR was employed to observe the structural modification of the samples. The spectra of the freeze-dried mucilage powder were obtained using an infrared Fourier transform spectrometer (model Vertex 70v, Bruker, Germany) with platinum ATR diamond f/ vacuum. The spectral range was from 400 to 4000 $cm^{-1}$ with 128 scans, resolution of 4 $cm^{-1}$, aperture setting 6 mm and acquisition rate of 10 Hz.

### 2.10 Statistical analysis

The results of the evaluated characteristics were submitted to Analysis of Variance (ANOVA) followed by the Tukey test to distinguish the treatments using on RStudio software (Illinois/NCSA) with a five-percent significance level. Extraction yield, total carbohydrates content, °Brix, pH, oil-holding capacity, water solubility index and viscosity were carried out in triplicate.

## 3 Results and discussion

### 3.1 Aqueous extraction

The crude efficiency for mucilage extraction was approximately 9.03%±1.22 for dry chia seed mass, 47.4%±2.12 dry psyllium husk mass, and 21.98%±0.69 for the combined mucilage.

The mucilage of chia seed has a strong attachment to the coat after water extraction making the separation an important factor in the efficiency of the process. Timilsenaa *et al.* (2016a) obtained 5.6% efficiency by conducting a lyophilization 1:20 (seed:water ratio) of soaked seeds with swollen mucilage and mechanical separation grounding into powder passing through 200 μm sieves. Muñoz *et al.* (2012) obtained 6.97% with 1:40 (seed:water ratio), 8.0 pH controled extraction, ten-hour drying at 50 °C, and mucilage separation from the seed rubbing the dried sample over a 40-mesh screen. Studies have successfully approached the efficiency factors (temperature, pH, water:seed ratio) with this particular paper focusing on the separation method. The 9.03% efficiency was achieved through the separation using cloth and a higher seed:water ratio (1:40). The highest ratio benefited the separation through a low thickness aqueous suspension. Additionally, the seeds were separated before the drying procedure and it is believed that extraction efficiency was higher due to the combination of previous separation of the seeds with higher seed:water ratio. Felisberto *et al.* (2015) achieved 7.86 g/100 g of chia seeds by separating the (1:40) aqueous solution with a brush depulper and a vaccum filtering before the drying stage.

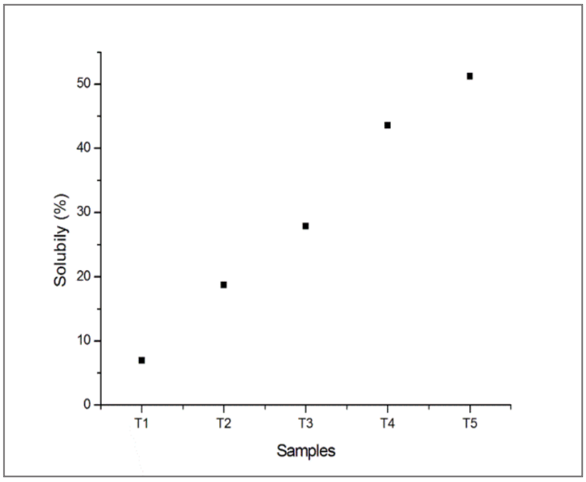
Despite the many resports on psyllium efficiency, only a few studies, such as Ahmadi *et al.* (2012), indicated 28% of filtrate dry matter for psyllium husk using hot water extraction. The method with more common application involves alkaline extractions followed by acid neutralization. No previous reports in literature are registered for the psyllium husk aqueous extraction combined with the chia seed; however, Equation 1 estimated a yield of 19.99%, which is numerically similar to the experimental result (21.98%). This study does not reveal any evidence of sinergic efficiency interaction between the both seeds during aquous extration using the studied conditions, although further experimental design studies would allow additional information by using controled factors such as pH.

### 3.2 Physicochemical and technological properties

The pH of the reconstituted mucilage ranged from 6.33 (T1) to 6.83 (T4) and the °Brix from 0.75 (T1, T2, and T3) to 1.0 (T6). Oil-holding capacity (OHC) ranged from 0.895 (T1) to 1.66 $mL_{oil}/mL_{sample}$ (T3). No significant difference between the samples was observed regarding those parameters. However the carbohydrate content of T5 (13.14 g/L) was significantly different from others where the values ranged between 7.37 (T1) and 8.68 g/L (T4).

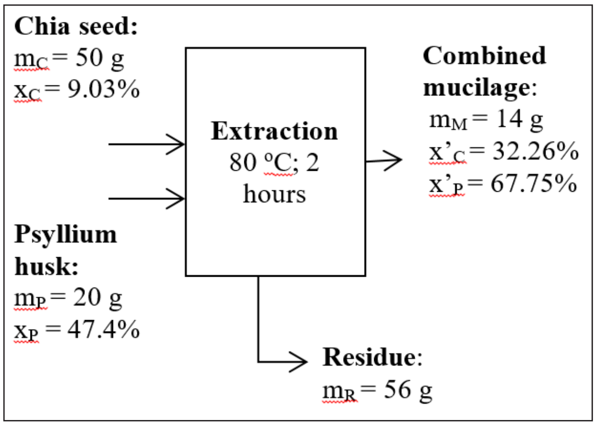
The water solubilization was measured using the WSI (Water Solubility Index) (Figure 1). The psyllium mucilage resulted in a solubility of 6.98% (T1) while the chia mucilage resulted in 51.26% (T5). By separating in two phases, gel and supernatant, psyllium uses most of its mucilage to form the gel chain while chia mucilage may loss the major part of it after centrifugation. The results of the blends indicated the expected behavior and the individual WSI revealed no evidence of a synergic effect of the blendings.

**Figure 1 –** Comparison of mucilage samples for the water solubility property



Considering that the WSI chart have a linear range ($R^2$=0.9913) between the blends, this study investigated whether the combined extration has the same behavior of the blended ones using the same mucilage proportion. In order to have it revealed, mass balance (Figure 2) was used to predict the amount of chia and psyllium mucilage on the combined mucilage. Accordingly, a sample containing a mixture of 32% C and 67% P was prepared for a triplicate test. The obtained WSI was 23.18%, close to the 22.63% found for the combined mucilage leading to the conclusion that the combined fraction is consistent with the individual mix at a proportion of 32% P and 67% C (between T2 and T3).

**Figure 2 –** Mass balance of combined extraction where mP and mC are the weight of psyllium husk and chia seed; xP and xC are the percentage of mucilage of raw material; mR is the residual weight; mM is the mucilage weight, and x'P and x'C are the final fraction of each mucilage

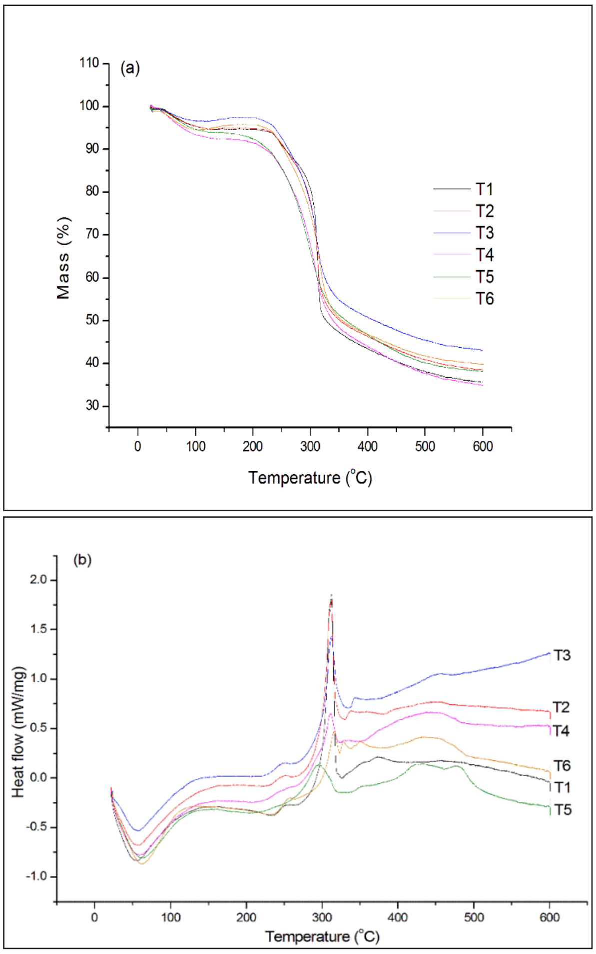


This result suggest that it is possible that the combined mucilage extraction have no interference on the solubilization of the individual mucilages. It is important and convenient when planning and developing a new product to consider an expected behavior for the mixtures.

## 3.3 Thermal characteristics

The TGA plot (Figure 3) of the samples revealed thermal effects similar to natural hydrogels reported in a few earlier studies (IQBAL et al., 2011a, TIMILSENA et al., 2016b). The observation of an endothermic loss had been previously reported due to the loss of absorbed moisture at a range of 50-100 °C. A possible explanation for the low temperature of moisture loss is the use of platinum crucible substituting hermetical sealing.

**Figure 3 –** (a) TGA and (b) DSC curves of mucilage samples



The average representative weight loss was 4.94% despite the significant difference between T3 and T4 (2.33% and 7.29%, respectively) (Table 1). Two stages of decomposition were observed for all of the samples. Generally, at stage one, the initial decomposition temperature (IDT) ranged from 142 to 211 °C and at stage two, the final decomposition temperature (FDT) ranged from 329 to 350 °C. Stage 1 resulted in a 45.63% weight loss in average, with an exothermic enthalpy alteration. This stage is responsible for the major breakdown of the polymer chain forming reasonably high molecular mass volatiles. Studies have reported an IDT of 220-280 °C and a FDT of 310-375 °C for chia, psyllium, and natural polysaccharides mucilage (TIMILSENA et al., 2016a, IQBAL et al., 2011a, IQBAL et al., 2011b). Chia mucilage results indicate the lowest weight loss of 41.33% and T6 indicate better stability with the major breakdown occurring at the highest temperature. The TGA peak previously observed in T4 was followed by the highest dehydration value. This study suggests that the amount of water in the polysaccharide chain could be responsible for the weakness of the main structure for exposing more boundaries compared to the others. The second decomposition stage was characterized by a decreasing curve up to the final temperature of 600 °C and a mean final weight of 10.75%. The char yield average was 38.68%.

**Table 1 –** TGA results of the mucilage samples

| Sample | Dehydration (%) | Stage 1 (°C) | TGA Peak* (°C) | Weight loss (%) | Stage 2 (°C) | Weight loss (%) | Char yield (%) |
|---|---|---|---|---|---|---|---|
| **T1** | 4.82 | 207-329 | 313.00 | 49.51 | 329-600 | 9.55 | 36.12 |
| **T2** | 4.78 | 207-330 | 312.67 | 46.44 | 330-600 | 9.83 | 38.95 |
| **T3** | 2.33 | 206-330 | 310.54 | 44.27 | 330-600 | 9.60 | 43.80 |
| **T4** | 7.29 | 165-330 | 302.04 | 44.55 | 330-600 | 12.89 | 35.27 |
| **T5** | 5.29 | 142-340 | 313.20 | 41.33 | 340-600 | 14.65 | 38.73 |
| **T6** | 5.14 | 211-350 | 315.17 | 47.66 | 350-600 | 8.00 | 39.20 |

*TGA peak was obtained by differentiate TGA curve.

Despite of the fact that psyllium and chia mucilage have thermal characteristics of hydrogels, it was possible to differentiate these materials by the degradation behavior on stage 1 and 2. Chia mucilage required more specific heat to keep the breakdown after stage 1 by the exothermic peaks observed. In addition, chia had the lowest weight loss (41.33%) at stage 1 followed by 14.65% loss over 340 °C showing that this mucilage has higher thermal stability compared to psyllium mucilage.

## 3.4 Viscosity

As a characteristic of non-Newtonian fluids, viscosity presented variations according to the shear rate applied (Figure 4). All of the treatments had mucilage indicating shear-thinning properties, attributed to the presence of high molecular weight materials; therefore, the apparent viscosity indicated a pseudoplastic behavior (LEÓN-MARTÍNEZ; MÉNDEZ-LAGUNAS; RODRÍGUEZ-RAMÍREZ, 2010).

**Figure 4 –** Viscosity curves of mucilage samples

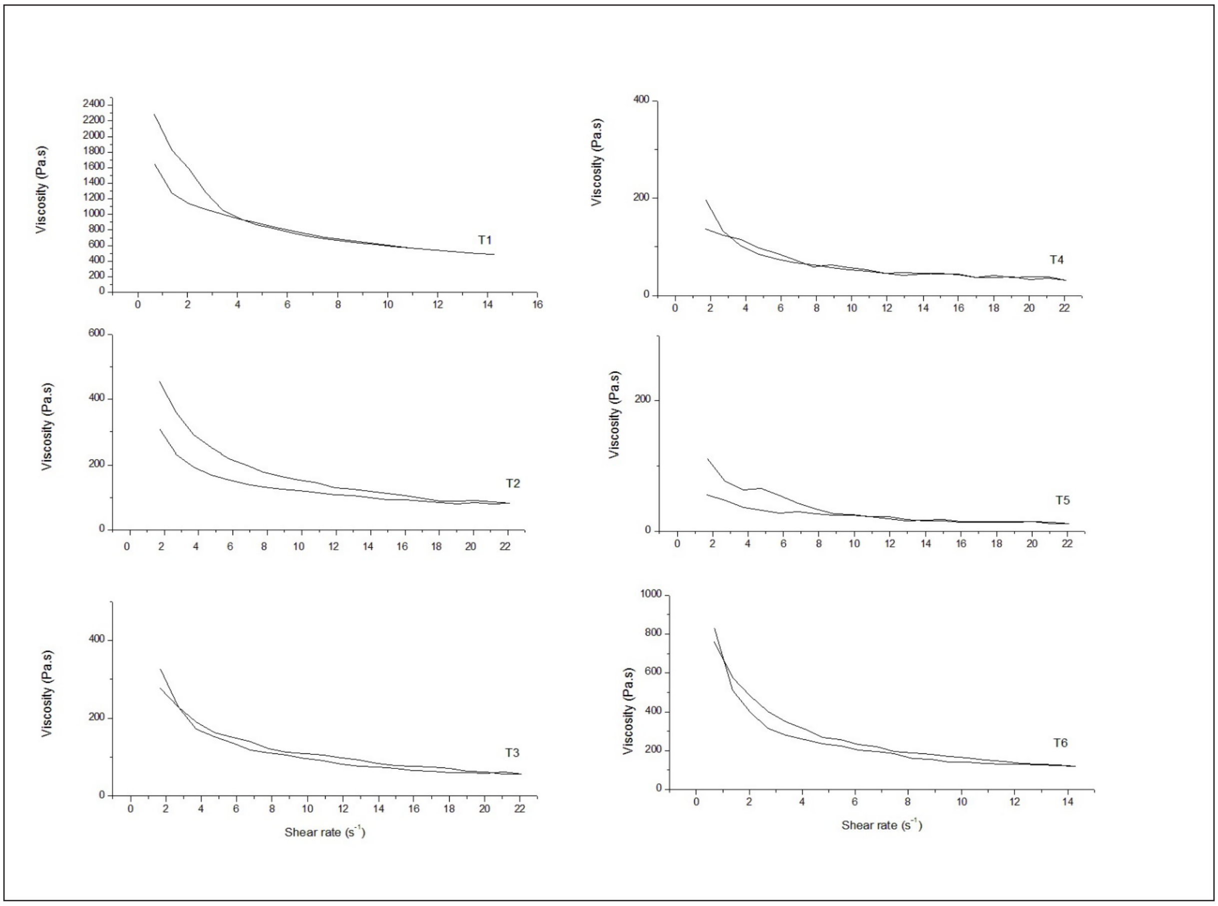
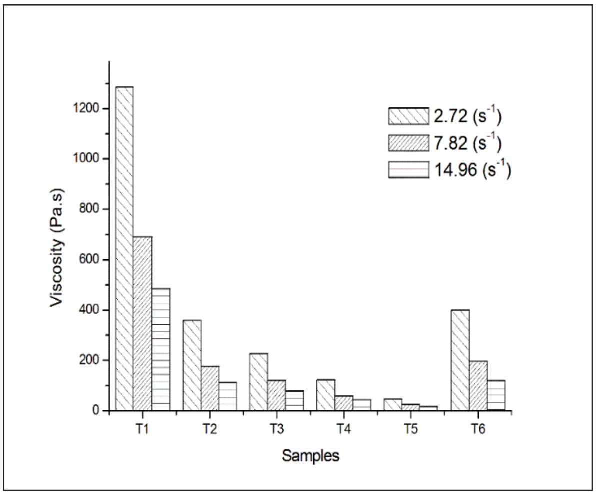


Figure 5 exhibits and compare three shear rate points (2.72 s-1, 7.82 s-1 and 14.96 s-1) of the viscosity curve. At 2.72 s-1, the viscosity has a downward trend once the chia mucilage concentration is raised (T1= 1285.86, T2= 360.27, T3= 228.47, T4= 123.02, T5= 46.87 Pa.s). This fact is also exhibited to 7.82 s-1 and 14.96 s-1 shear rates. Beyond that, T6 presented unexpected behavior revealing the highest value of the blends (401.28 Pa.s). According to mass balance, the expected results were meant to be between T2 and T3. This is an indication that the aquous combined extration acts differently than a simple mucilage mixing.

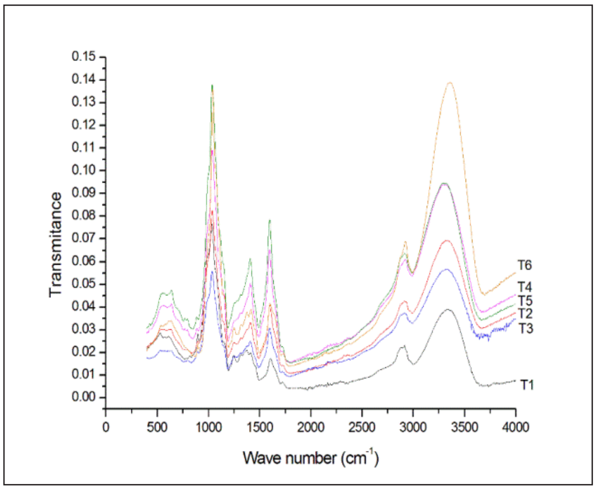
**Figure 5 –** Viscosity comparison of mucilage samples of three different shear rates



## 3.5 ATR

Figure 6 illustrates the ATR spectra of the samples. Previous studies had obtained similar spectra for psyllium and chia mucilage. By comparing the samples, it was observed that the similar broad bands ranged between 3358 to 3307 cm$^{-1}$, representing the hydroxyl (-OH) stretching of the gross carbohydrates; 2928 to 2886 cm$^{-1}$, the C-H stretching of the aromatic rings and the methyl group (CH$_3$); 1724 to 1726 cm$^{-1}$, the C=H stretching vibration of carboxylic acid; 1598 and 1420 cm$^{-1}$, and the symmetric stretching of carboxyl group (-COO-) ion present in uronic acids. The bands at 1750 cm–1 and 1155 cm–1 may represent the bending vibration of C=O and C-O-C of the pyranose ring. At 1038cm$^{-1}$ can be related to C-O-C stretching of 1→4 glycosidic bonds and C-O-H bending, considered a characteristic of polysaccharide compounds. The band at 844 cm$^{-1}$ may represent the β-nanomeric C-H deformation and glycosidic linkages attributed to glucopyranose and xylopyranose units (CERQUEIRA et al., 2011, TOGRUL; ASLAN, 2003, TIMILSENA et al., 2016b). The difference between the samples occurred at the 700 cm$^{-1}$ range, representing cis C-H out-of-plane bends, at 1250 cm$^{-1}$ (C-O- stretching), 1377 cm$^{-1}$ (-CH3 symmetric bend), 821 cm$^{-1}$ (C-O out-of-plane bend) and 1653 cm$^{-1}$ (>N-H of secondary amine) (TIMILSENA et al., 2016b, COATES, 2006, TIPSON, 1968).

**Figure 6 –** ATR spectra of the mucilage samples



In general, sample bands in certain regions emerge or become more visible with the addition of the different mucilage. For example, the bands of 770 cm$^{-1}$ and 795 cm$^{-1}$, where T5 and T4 present peaks, T3 and T2 have a subtle increase and T1 revealed no evidence of a peak. Figure 7 illustrates results from 400 to 900 cm$^{-1}$ ATR spectra. Sample T6 did not present the same tendency of the mixtures having indicated a similar behavior to the psyllium mucilage (T1) in some regions. In addition to not seem to have the above-mentioned peak (770 cm$^{-1}$ and 795 cm$^{-1}$), the similarity between samples T1 and T6 appear in peaks at 1248 and 1377 cm$^{-1}$ (Figure 8). The remaining samples (T2 and T3) revealed only one increase soften with the increases in the chia mucilage concentration (T4 and T5). Differences between T1 and T6 molecules were found in bands 638 cm$^{-1}$, 821 cm$^{-1}$ and 1653 cm$^{-1}$. Assuming that the molecular structure, among other factors, influences the properties of a polymeric gel, the ATR spectra may explain the T6 viscosity behavior above expectation since its conformation is similar to T1 (highest viscosity values).

**Figure 7 –** ATR spectra of the region 1 (from 400 to 940 cm$^{-1}$)

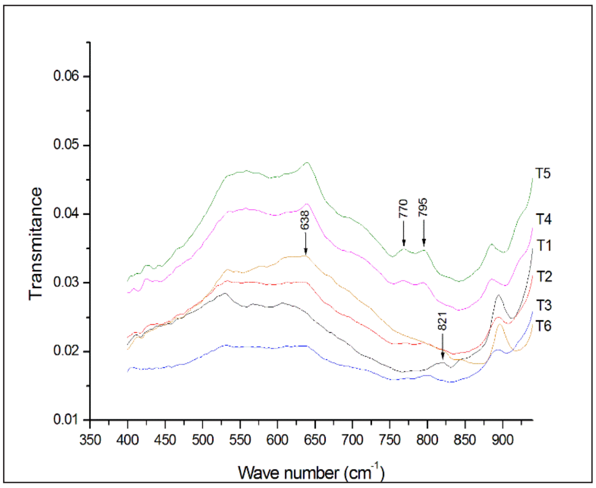
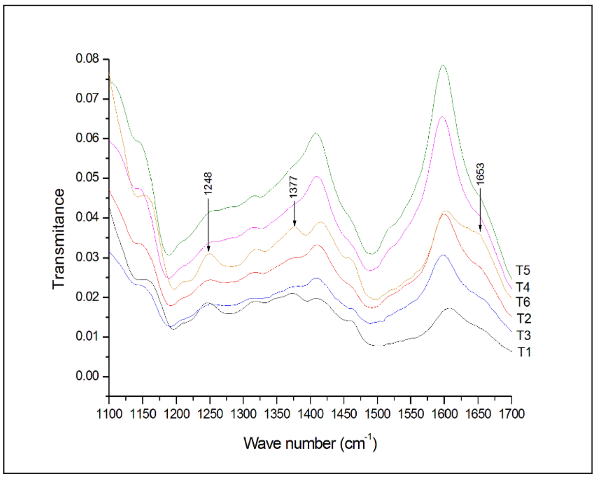


**Figure 8 –** ATR spectra of the region 2 (from 1100 to 1700 cm$^{-1}$)



## 4 Conclusion

It is important for the industry to understand the interactions among ingredients in order to achieve optimum production or even develop a new product. This study revealed that mixing chia and psyllium mucilage results in a different behavior when compared to the combined extraction since the viscosity results were higher than expected. Among the blends the T6 presented the highest viscosity with 401.28 Pa.s. Also, T6 indicate better stability during the TGA analysis. For this reason, the combined mucilage is the highlight of the work. As the industry seeks to leverage factors, such as thickening and the absence of artificial ingredients addition, combined extraction processes may constitute an interesting alternative. The ATR spectra was an important instrument to investigate the causes of the increased viscosity for revealing on a molecular level the similarities of the highest viscosity samples structures.

## 5 Acknowledgements

The authors are thankful to the Brazilian agencies CAPES, CNPq, and Fundação Araucária for their financial support of this work.

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